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CLINICAL AND ANATOMIC RELATIONS IN CHRONIC NEPHRITIS *

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NEW YORK

The pathologist who is also a clinician finds himself constantly in a state of disillusionment. He finds that many conventions of clinical medicine and anatomic facts do not always coincide, not in the broader sense of diagnosis, but in the finer interpretation of clinical data by pathologic diagnosis and vice versa. After years of such attempts at interpretation he becomes painfully aware that pathology, after all, does not explain everything in clinical medicine, and that in order to bring current medical lore and pathology into closer relationships, many readjustments are necessary. There can be only two such reasons why this should be: either clinical medicine is wrong insofar as clinical data have not been sufficiently "checked up" with anatomic facts in the past; or our pathology is wrong insofar as these facts have been falsely interpreted in terms of clinical medicine. Both these factors are at fault, but the clinician is more to blame because he shifts the entire burden of responsibility for the interpretation of clinical data on the shoulders of the pathologist, who is obviously in no position to interpret them. The pathologist, too, is at fault, not so much because he knows little of clinical medicine, but because in his enthusiasm for pure morphology he is apt to relegate pathogenesis and pathologic physiology into the background of his consciousness. No one who has pondered over these things can fail to realize that a large field for investigation lies in the closer correlation between clinical medicine and pathology. Tradition must be tested by the touchstone of critical observation.

These reflections are the result of studies on nephritis [1] recently published by the writer. In attempting to reconcile traditional clinical medicine with the material obtained at the autopsy table he found that, except with limitations, certain supposed varieties of clinical

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* Read before the American Association of Pathologists and Bacteriologists, New York City, April, 1920.

1. Moschowitz, E.: Hypertension; its Relation to Arteriosclerosis and Nephritis and Etiology, Am. J. M. S. **158**:668 (Nov.) 1919.

nephritis did not correspond with findings at necropsy; and, on the other hand, certain pathologic lesions afforded little more than a pure guess in attempting to predict clinical phenomena. In attempting to interpret these discrepancies the writer drew the following deductions: (1) That in the majority of instances what the clinician means is not "nephritis" but renal insufficiency. In other words, the diagnosis must be couched in terms of function rather than in those of anatomy. (2) That the large group of the so-called "clinical varieties" of chronic nephritis are remarkably indefinite things and have no precise morphological substratum. (3) That so-called various types of chronic nephritis are not end products but stages in a well defined pathogenesis.

1. *Chronic Nephritis Versus Renal Insufficiency.*—Before discussing the nature of these terms certain definitions are in order. The conception that chronic nephritis is a reaction to an injury has not been superseded. This is represented by more or less permanent changes in the organ in which the development of new connective tissue is the predominant lesion. All of the various components of the kidney structure partake in this change. There is no need to describe here in detail the histological characters of chronic nephritis. Briefly, the changes are the following:

(a) The glomeruli: These show a connective tissue change beginning with an increase in the intravascular and the extravascular endothelium [2] of the Bowman capsule, progressing to a connective tissue transformation of these cells, a thickening of the walls of the capillaries, and eventually partial or complete hyaline degeneration of the entire glomerulus, sometimes with calcification. Here I wish especially to emphasize that the process I have briefly described is wholly a vascular change and represents in miniature the same process as that termed "arteriosclerosis" affecting the larger vessels. The significance of this conception will be dwelt on later.

(b) The Epithelium: The epithelium may show no changes whatever. In the more advanced stages, it reveals degenerative changes, often fatty. In advanced nephritides much of the epithelial structure is deformed and destroyed by the encroaching widespread new connective tissue formation. With the destruction of some of the tubules compensatory changes occur in those that remain. There is an apparent proliferation with the formation of adenoma-like masses that

2. Speculation has been abundant as to whether the lining cells of the Bowman capsule are endothelium or epithelium. Certainly these cells possess all the potentialities of endothelial cells and none of epithelium. In studies upon "Angiogenesis and Ossification (Bull. Johns Hopkins Hosp. **27**:71 (March) 1916), I arrived at the conclusion that the function of endothelium was not specific, but that it could assume all the characters of any cells of mesothelial origin.

contribute greatly to the glandular appearance so often present in advanced nephritides.

(c) The Interstitial Tissue: The changes in the interstitial tissue begin with a slight infiltration of round cells in the cortex, usually in the neighborhood of diseased glomeruli. This is, in all probability, the result of multiple minute infarcts, both of the glomeruli, the arteriae rectae and the larger vessels. In the more advanced stages the round-celled infiltration is more diffusely distributed between the tubules, producing a swollen and whitish appearance of the cortex. In the advanced stages this infiltration has been converted into frank fibrillar connective tissue of varying density. With the scar-like contraction of this tissue and the corresponding atrophy of the intervening parenchyma, there result a thinning and irregularity of the cortex, pitting and indentations on the surface, and, finally, when this scarring involves the organ diffusely, we find the so-called "contracted" kidney. This is the only true end result.

(d) The Blood Vessels: This obviously excludes the glomeruli. In the early stages, the changes may be few or none. In the later stages, the changes correspond to those ordinarily seen in arteriosclerosis of the larger vessels. These changes are too familiar to be described here. In addition to these changes, a passive congestion is an exceedingly common accompaniment, even without a frank valvular lesion of the heart. The significance of this lesion I shall also dwell upon in a subsequent portion of this paper.

Renal insufficiency is an exceedingly complex thing and is consequently not easy to define. The function of the kidneys as a whole, to say nothing of its separate components, is by no means a settled subject: nor are we quite sure to what extent some of the functions that have been attributed to the kidney are purely renal or of extrarenal origin. These are the reasons why it has been difficult to gauge the value of certain functional tests currently employed to test renal insufficiency, so that one must approach the study of renal insufficiency in an extremely critical frame of mind. At all events, some of these tests have proven beyond doubt, by the results of operation, by anatomical findings and otherwise, that they are of value in prognostically estimating renal insufficiency.

I shall not enter at present into a discussion of the relative values of any of these tests. This subject is still in the formative stage and is rich in speculation. In reviewing this work, however, certain facts stand out prominently. 1. That no single test is an adequate guide to renal function, but that a larger number in various combinations must be performed in order to gain an even fair perspective of the renal function. In other words, it is not function but functions that must

be determined. 2. That a single determination of one or various tests gives little prognostic information and indicates only an approximate determination of renal insufficiency at the time at which the tests are performed. To be of any constructive value these tests must be repeated during the entire course of the illness. 3. A corollary of the previous observation: That these tests are profoundly influenced by extrarenal factors. Among the most important of these factors are the following: (a) diet, especially fluid intake; (b) the condition of the patient; whether he is afflicted with a coexisting disease, etc.; (c) changes in the function of some of the other organs, the liver for instance, due either to the direct result of the kidney disease or to circulatory changes consequent upon the renal malady; (d) circulatory insufficiency. This is, in my estimation, the most important factor.

These observations lead one to suspect, what, indeed, proves to be true, that renal insufficiency cannot be estimated, except to a limited extent in terms of the amount or degree of anatomic injury to the kidney. I have for years attempted to correlate the extent and variety of damage of either of the glomeruli and tubules with the clinical findings expressive of disordered renal function, and have thus far failed to determine any precise relationship. In the vast majority of instances, it is true, a lesion of the kidney was found, but one could rarely predict clinically, except within certain limitations, what the extent and variety of the lesion would prove to be. Indeed, it is perfectly possible to find kidneys with little or no injury to the parenchyma, although all the evidence during life indicated impairment, even severe, of the renal function. On the other hand, badly damaged kidneys are often found at autopsy, although the clinical findings during life were normal or nearly normal. This is especially evidenced in the so-called "secondarily contracted kidney" or the "arteriosclerotic kidney" of Sutton and Gull. During life patients who harbor such kidneys reveal little in their urine, except occasionally traces of albumin and a low specific gravity. In such patients Larkin and Levy [3] found normal nitrogen figures in the blood, except just before death. Nevertheless, such kidneys are frequently present as accidental findings; the patient does not die in uremia, as we should expect, but from an intercurrent infection, cardiac failure, arteriosclerosis, etc. These facts explain the futility of all attempts to explain symptoms, urinary findings and other tests of renal insufficiency, including hypertension, etc., on a mechanistic basis. The most recent attempt has been that of Elwyn,[4] but Christian [5] in an admirable critique reveals the fallacies of Elwyn's assumptions.

3. Larkin and Levy: International Clinics, 1918, ii, 26.
4. Elwyn: Am. J. Urol. **3**:47, 1919.
5. Christian: Progressive Med. **24**:119, 1919.

To discuss all the possible relationships of each of the numerous clinical findings and tests for renal function at present at our command would be futile in the light of the inadequacy of our present knowledge of the significance of these tests. I shall, in this place, merely discuss three of the most important, viz., albuminuria, hypertension and albuminuric retinitis.

Albuminuria.—In a previous paper I have already discussed the significance of this symptom and its relation to anatomic damage to the kidney. I tried to show that neither its presence nor its quantity is at all commensurate with the variety and degree of damage to the kidney. Albumin may be present in considerable quantities even when the kidneys are entirely normal, as in orthostatic albuminuria, or in cases of circulatory insufficiency due to valvular heart disease, disappearing when sufficiency has been restored. In both instances, especially the latter, the presence and disappearance of the albumin cannot be explained on other than purely physiologic grounds. On the other hand, albumin may be absent or practically absent, even when the kidney has suffered extensive damage, as in the so-called "contracted" kidney. It apparently makes little difference whether the glomeruli or the tubular portions are involved or both; the presence or quantity of albumin bears little relationship to the amount or degree of damage to the kidney.

I am willing to concede, indeed, I have demonstrated this to my own satisfaction, that albumin is usually proportional to the extent and degree of the tubular degeneration associated with many infections, (e. g., typhoid fever, pneumonia, subacute bacterial endocarditis) the albumin resulting from cell autolysis or increased filterability, but I do not believe that the changes in the tubular epithelium are to any great extent a determining factor in causing albuminuria in the other nephritides, especially of the chronic variety.

These observations leave us no ground for assuming that the kidney is merely a filtering organ, an assumption that physiologists have already amply disproven. The presence of albumin is determined by other factors than mere damage to the glomeruli or the tubular cells. Pressure and volume changes within the blood vessels, the composition of the blood, especially of the threshold bodies [6] and as yet little understood colloid changes in the plasma are equally, if not more, important factors in causing albuminuria, as are pathologic changes within the kidney.

I am fully aware that albumin is probably not the main or even the earliest evidence of renal insufficiency. As Myers and Kilian [7]

6. Cushny: The Secretion of Urine, London, 1917.
7. Myers and Kilian: Am. J. M. Sc. **157**:674, 1919.

have shown, diminished phthalein and increased blood urea and creatinin figures may be found even when albumin is absent from the urine. I simply mean to convey that albuminuria bears no consistent relation to the amount or degree of pathological changes within the kidney, that is, so far as our microchemical methods permit us to determine.

Hypertension.—In the paper already mentioned I submitted the thesis that hypertension was not a consequence of nephritis or arteriosclerosis, but that it was a primary compensatory phenomenon due to unknown causes, and that the nephritis and arteriosclerosis represented secondary changes due either to the hypertension itself or to the cause or causes of the hypertension. I shall not here detail the lines of reasoning that led me to this conclusion; I shall simply say that subsequent observation and reflection have confirmed me in this belief. My conception expressed in terms of clinical medicine is the following: The vast majority of the nephritides or cardionephritides began clinically as cases of so-called "essential" hypertension; i. e., the patient reveals on physical examination a greater or lesser degree of hypertension, greater on the diastolic scale, with or without cardiac hypertrophy, but without evidence of renal insufficiency as apparently evidenced by the absence of albumin or casts. I am not aware at this time of studies that show the status of other tests of renal insufficiency in clinical cases of essential hypertension, but I would not be surprised if such tests showed evidences of renal insufficiency, even though albumin were absent. In any event, if one follows such cases, as I have done, one finds generalized changes occurring in the tissues of the body, especially in the arteries, notably the retinal, the coronaries, the cerebral and the renal.

If arteriosclerosis, the albuminuric retinitis and nephritis are analyzed in their morphologic and genetic aspects, no one, I believe, can escape the conclusion that they are all manifestations of one and the same lesion, a primary vascular productive inflammation affecting all the blood vessels from the capillaries to the aorta. In the kidney, as I have described, and as all authorities are agreed, the primary change is in the glomeruli. This represents the type known as "glomerulonephritis." As the process advances, the larger vessels become affected. All subsequent lesions in the kidney may be traced morphologically from these primary changes in the blood vessels of the organ. Owing to the destruction or impoverishment of the blood supply of focal areas in the kidney, smaller or larger infarcts occur which in turn produce new connective tissue, first in the form of round cell infiltration, and later in the form of fibrillar and hyaline connective tissue. Much of the degeneration of the epithelium can be explained

by the same pathogenesis, although some of it is probably due to waste products from the altered metabolism. The disappearance and malformation of the tubules are accounted for in large measure by the destructive effects of the new connective tissue transformation.

The kidney in chronic nephritis may, therefore, be conceived as a tissue filling the interstices of a framework of blood vessels, depending on the integrity of these blood vessels for its preservation and function. When this framework is injured, the organ, like a house, will fall to pieces. This is the reason why I agree with those who hold that chronic nephritis is a primary vascular disease, e. g., Jores. *Chronic nephritis and arteriosclerosis, therefore, are, to my mind, one and the same lesion.* I believe it is incorrect to view arteriosclerosis, either clinically or pathologically as a disease affecting the larger vessels only. It affects the entire arterial system, from the capillaries to the aorta.

The lesions in chronic nephritis, as a result of this local arteriosclerosis, may be viewed as representing a continuous series of granulomatous changes, and all phases of the morphology may be explained in the light of such changes. The various lesions encountered in chronic nephritis are subject to the identical laws of morphology as apply to granulomatous changes in other tissues from whatever causes. Thus the changes in the kidney are entirely analagous to those witnessed in the various stages of cirrhosis of the liver, or of chronic oöphoritis or in an ordinary granulating wound.

Retinitis.—A more readily grasped analogue, which few appreciate, is the complete identity even down to the minutest details between the lesion of chronic nephritis and those of albuminuric retinitis. Accept any description of the pathological lesion in retinitis, that of Collins and Mayou [8] for instance, and the lesions in both nephritis and retinitis are paralleled in every detail, both in morphology and in the order of their appearance. Indeed, these authors admit that the lesions "are in reality degenerative processes secondary to changes in the retinal vessels." This analogue is, I believe, a strong support for the thesis that the nephritis is not primary but a result of the hypertension: for this type of retinitis as far as my observation leads me to believe, is never present unless hypertension is or has been present, and if the retinitis is sequential to the hypertension or to the cause or causes of the hypertension, it seems to be a logical deduction that the nephritis, which is the identical lesion, cannot but be a consequence as well. It would be an interesting study to see how far the nephritis and the retinitis parallel each other in gravity.

8. Collins and Mayou: Pathology and Bacteriology of the Eye, Philadelphia, 1911.

Depending, therefore, on which organ is most affected a certain clinical picture is developed, which may be that of generalized arteriosclerosis, a localized arteriosclerosis (cerebral arteriosclerosis) apoplexy, aortitis, coronary sclerosis (resulting in chronic myocarditis), and finally chronic nephritis. The patient usually presents himself at a time when the more advanced stages are manifest, but if we observed the real beginnings of the disease, a circumstance, as Mackenzie emphasizes, of fundamental importance in the biological study of disease, I believe that practically all such cases would have revealed the clinical syndrome of "essential hypertension." Certain it is, that the presence or degree of hypertension bears little or no relation to the presence or degree of damage done either upon the arteries or the kidney. Just as we see hypertension often associated with little or no disease of the arteries and profound disease of the arteries (decrescent type of Allbutt) associated with no elevation of blood pressure, so we see increased blood pressure where the kidneys are normal or practically normal[9] (Hirschfelder,[10] Mosenthal[11]) while, on the other hand, normal blood pressure may be present in patients whose kidneys are profoundly diseased. The latter phenomenon is seen not uncommonly in association with the senile or "arteriosclerotic" kidney, a form which corresponds, in my belief, to the decrescent type of arteriosclerosis and is, moreover, indistinguishable morphologically from the contracted kidney resulting from a progressive glomerulonephritis.[12] Indeed, all attempts to correlate these two phenomena have thus far failed, either when we conceive of the destructive process within the organ as a whole, or analyze it in terms of the different components of the kidney structure. This is one of the main arguments I have adduced to support the belief that the hypertension is the primary and the arterial and kidney lesion the secondary phenomenon. Genetically, the hypertension certainly preceded the signs of nephritis in a very large percentage of all cases, as any clinician with experience can testify. Moreover, this belief is supported by the fact that all other chronic nephropathies, morphologically easily definable

9. I have recently acquired two such kidneys in patients in whom the systolic blood pressure was over 200. These will be reported later.

10. Hirschfelder: Diseases of the Heart and Aorta, Philadelphia and London, 1918.

11. Mosenthal: Medical Clin. N. America, July, 1917, p. 101.

12. Vide MacCallum (Textbook of Pathology, 1917): "The gross appearance of such a kidney (arteriosclerotic) differs in no way, therefore, from the end result in a protracted glomerulonephritis (the so-called secondarily contracted kidney), and in all probability something similar may result if the destruction be severe enough and time be given in the case of the tubular and interstitial forms. As a matter of fact, except perhaps through the presence of the strikingly thickened blood-vessels, it is difficult if not impossible to distinguish microscopically the types of ultimately contracted kidney."

from that which I have described, amyloid, subacute bacterial endocarditis, chronic surgical lesions, experimental destructions, congenital cystic kidneys, parenchymatous nephritis,[13] etc., are never associated with hypertension.

The reason why we cannot predict the variety or degree of changes, in other words, the "type" of kidney that we shall find from clinical findings alone, lies, I believe, in the fact, that these changes or "types" do not represent end products, but only phases of a definite progressive morphologic pathogenesis, so that the "type" of kidney found at autopsy does not represent in any sense the final result, transcribed in reports as the "cause of death", but rather, that particular stage of the disease in which the patient happened to die, whether from an apoplexy or coronary thrombosis or uremia. The nephritis is therefore a glomerular nephritis in the beginning; a diffuse or granular nephritis, which may be white or red or mixed, large or small, in the next stage; and a contracted kidney in the final stage. The only stage that can on morphological grounds be consistently defined as an "end product" is the final stage of such a pathologic process, the so-called "contracted" kidney. Genetically, therefore, it is reasonable to suppose that in the so-called "essential" hypertensions the kidneys present little damage, limited to the glomeruli where the early changes in chronic nephritis originate; while in the hypertensions in which renal functional tests show a profound insufficiency the damage to the kidney is diffuse and extreme. I cannot state this with any reasonable degree of exactitude, because both my experience and the reported observation are inadequate. The difficulty is that patients with "essential" hypertension do not often die in that stage.

The evidence which I have presented is sufficient, I believe, to demonstrate that it is extremely hazardous to predict the amount and degree of damage in the kidney by clinical findings alone. It now remains to determine, if possible, the factor or factors that cause this discrepancy. Not being anatomic, the factors must be physiologic. This is necessarily so, because productive inflammations never completely disappear.

I am convinced that by far the largest factor or rather the largest common denominator of the entire problem is circulatory insufficiency. No one who has seen anything of the clinical phases of "chronic nephritis" can fail to be impressed by the dominating rôle of circulatory insufficiency from the very beginning of the malady to the end. In the earlier stages, whether we view it in the light of a compensatory mechanism or not, this insuffiicency is manifest in a more or less persistent hypertension, with a resulting hypertrophy of the left ven-

13. Epstein: J. A. M. A. **69**:444 (Aug. 11) 1917.

tricle. In the further course of the disease, other evidences of a progressive circulatory failure arise. These are increased rapidity of the heart action, disturbances in rhythm (gallop rhythm, extrasystoles, auricular fibrillation) and signs of stasis (enlarged liver, hypostasis, edema of the ankles, etc.). If coronary sclerosis occurs, with resulting fibrotic changes in the myocardium due to the blocking of the terminal branches of the coronary vessels, all signs of a real anatomical myocarditis become manifest. In addition to those already mentioned, two very important signs now appear. The first is a lowering of the blood pressure; the second is a change in the electrocardiographic tracing, described by Oppenheimer and Rothschild [14] as "arborization block." During this stage of the illness, which may be described as the middle phase, the patient is responsive to digitalis, and this, despite the fact that the hypertension may not be influenced to a considerable degree; so that by the continuous administration of this drug we are able to keep the subjective and objective evidences of circulatory insufficiency at a certain level; the circulation, in other words, may be said to be compensated. When the signs of circulatory insufficiency increase (decompensation) an increase in the quantity of digitalis combined with bodily rest usually suffices to restore the patient to his previous condition. Finally, it is noted that the attacks of decompensation increase in severity, frequency and duration. All the signs of a grave circulatory insufficiency ensue; the patient no longer responds to circulatory "stimulants" and unless his life is cut short by an apoplexy or an attack of angina pectoris, he dies from a pure circulatory failure or from uremia.

I have used the term "circulatory insufficiency" in a rather broad sense, for I am fully aware that this term comprises many elements aside from blood pressure, changes in rhythm, etc., such as myocardial tonus, blood velocity, volume flow and the viscosity of the blood, but until further investigation enlightens us on the relative importance of these elements in bringing about circulatory insufficiency, we must depend on a clinical concept of this phenomenon.

At all events, the similarity of the circulatory insufficiency noted in the nephritides associated with hypertension and that witnessed in frank valvular disease of the heart is striking. They parallel each other closely not only in signs and symptoms but also in the sequence of clinical events and their response to treatment. The same laws of cardiodynamics apply to both. Furthermore, there is one lesion representing circulatory insufficiency common to both, namely, congestion (Stauung) of the internal viscera. This is shown in brown induration of the lungs, nutmeg liver, chronic congestion of the spleen and

14. Oppenheimer and Rothschild: J. A. M. A. **69**:429 (Aug. 11) 1917.

cyanotic kidneys. These lesion I have found exceedingly common in autopsies upon cases of "chronic nephritis." The point I wish to bring out is that clinically tests for renal insufficiency are usually coordinate with evidences of circulatory insufficiency, and that the functional state of the circulation has a greater effect in increasing or decreasing the evidences of renal insufficiency than perhaps any other factor.

Not all the conventional functional tests are affected in this manner; I refer especially to quantity of urine, albumin, phthalein, urea and nonprotein nitrogen. Creatinin, uric acid and salt are not so readily affected. The figures rise and fall depending on the condition of the circulation. When the heart action is good, the figures approach normal, and conversely, when it is bad; just before death the figures show the widest variation from the normal.

These observations, it seems to me, receive a strong corroboration from what we find in cases of circulatory insufficiency due to pure valvular disease in which a genuine anatomic nephritis can definitely be excluded. We all know that in mild compensated valvular lesion of the heart, tests for renal insufficiency usually reveal normal findings. We also know that when signs of myocardial or, perhaps, better said, circulatory insufficiency develop, signs of renal insufficiency manifest themselves. There is oliguria; albumin and casts appear, phthalein excretion is reduced, and the blood urea nitrogen figures are above normal. When compensation is restored by rest and digitalis, these promptly disappear. Very often patients exhibit evidences indicating a combination of cardiac and renal disease so that there may be doubt as to how far the malady is cardiac or renal. With the improvement in the condition of the circulation, all signs of renal impairment disappear. If they do not completely disappear we may accept this as evidence that either complete compensation is no longer possible, or, less likely, that an associated productive renal lesion is present.

Observations of Rowntree, Geraghty and Fitz[15] are extremely important as confirming this view. They found in moderate experimental congestion of the kidney, that the excretion of salt, iodid and lactose may be interfered with. The excretion of the phthalein may be normal, but if the congestion is extreme, it may be decreased. "Therefore, in experimental congestion, functional pictures indistinguishable from those of mild or advanced nephritis without cardiac decompensation have been found. The gradual amelioration of experimental congestion is accompanied earliest by an increased phthalein output, later by improvement in salt and iodid excretion, and lastly by a slight amelioration of the delayed lactose excretion." Rowntree

15. Rowntree, Geraghty and Fitz: Arch. Int. Med. 2:258 (Aug.) 1911.

and Fitz [16] found that clinically cases of passive congestion behaved in a similar way.

These observations, it seems to me, render the examination for renal insufficiency of little value when made during the attacks of decompensation. For purposes of prognosis such tests, as Christian insists, should be made in the interval.

Anatomic Varieties of Chronic Nephritis.—From the foregoing discussion it may be gathered that clinical findings offer little or no basis for furnishing a classification of the forms of chronic nephritis that I have described. This form has received a host of names in textbooks; glomerulonephritis, productive nephritis, diffuse nephritis, interstitial nephritis, arteriosclerotic disease of the kidney, arteriocapillary fibrosis, primary and secondary contracted kidney, etc. Each of these names conveys some aspect of the pathology, but in order to obtain an adequate concept, which must include the origin, spread and progress of the disease, the structures involved and the physical characters of the diseased organ, all of these adjectives would be necessary as prefixes. No single name or group of names can suffice. It seems to me that the term "arteriocapillary fibrosis," devised by Gull and Sutton, is the most applicable. However, it may be taken for granted that none of the methods of clinical examination at our command can enable us to foretell whether we shall find such a kidney in the stage of a glomerulonephritis or that of the large or small red kidney, or the granular kidney or the contracted kidney. All that we have the right to say is that we shall find a kidney of this particular type. It has seemed to me from the few observations I have made, that the longer the duration of the disease, the more progressive the lesion. This is certainly a reasonable expectation.

The one unfailing clinical finding associated with this form of nephritis is hypertension. No other nephropathy is associated with this phenomenon. There are two exceptions to this rule. (1) Cases that have developed coronary sclerosis with symptoms of angina pectoris. The resulting fibrous myocarditis causes such an insufficiency of the heart muscle as to apparently neutralize the increased peripheral resistance. Anamnesis of such patients reveals the interesting fact that previously, perhaps a year or two ago, the blood pressure had been considerably elevated. I have a number of records of such cases. I have seen one case of angina pectoris in which this decrease occurred under my observation. In another, the associated interstitial myocarditis was proved by autopsy. (2) Cases of "decrescent" nephritis. I call this variety decrescent because it clinically corresponds to the decrescent arteriosclerosis of Allbutt. These patients reveal at

16. Rowntree and Fitz: Arch. Int. Med. **2**:258 (Aug.) 1911.

necropsy extensive general arteriosclerosis and the so-called "arteriosclerotic" or "senile kidney" or the "arteriocapillary nephritis" of Sutton and Gull. As Allbutt has pointed out in connection with the decrescent arteriosclerosis, so in this form of nephritis there is no hypertrophy of the left ventricle, and it is the exception rather than the rule for these patients to die from the causes common in the diseases of the hypertensive type.

The other sign characteristic of the hypertensive type of nephritis is an albuminuric retinitis. As far as I am aware, this type of retinitis is practically always associated with hypertension either present or past.

There are, however, other chronic nephropathies with different genetic and morphologic characters of their own, which while offering many of the signs and symptoms of the hypertensive type, yet possess certain symptoms and findings sufficiently characteristic to render them clinical entities as well. Most of the tests for renal insufficiency are common to both.

I shall exclude for obvious reasons the chronic surgical and specific nephropathies.

There remain for discussion the following varieties of chronic nephritis:

1. *Parenchymatous nephritis or nephrosis (Epstein).*—Although pathologic studies of this variety of nephritis are still lacking, it seems necessary, because of its clear cut clinical characteristics, to recognize this malady as of an entirely different type from the foregoing. This type of nephritis is a disease of youth or middle age. The urine is usually diminished in amount, rich in normal solid constituents and in albumin and casts. Edema and anasarca are early and prominent symptoms. But the most characteristic findings, according to Epstein, are the impoverishment of protein in the blood plasma, with an increase in the globulins, and a relative or absolute increase in the lipoid and cholesterol bodies. Epstein has obtained excellent results in this variety of nephritis by a diet high in protein and low in fats.

2. *Chronic glomerulonephritis associated with subacute bacterial endocarditis (Streptococcus viridans).*—While in the literal sense this lesion is not a chronic one, except in the healed stage, it is included in this discussion. This variety was first described by Löhlein [17] and more recently by Baehr.[18] The lesion according to Baehr is characteristic of subacute bacterial endocarditis and occurs in no other hematogenous bacterial infection. The lesion is a progressive one, is limited strictly to the glomeruli, and consists in swelling, degeneration and desquamation of the cells of Bowman's capsule, later followed

17. Löhlein: Ueber die entzündlichen Veränderungen der Glomeruli be i menschlichen Nieren und ihre Bedeutung für die Nephritis, Leipzig, 1906.
18. Baehr: Proc. New York Path. Soc., December, 1911.

by fibroblastic changes in the capsule and eventually (in the healed stages) by hyalinization of this mass. The new connective tissue formation never proceeds beyond the confines of the glomerulus. This lesion affords to my mind a convincing refutation of the argument of those who regard a glomerulonephritis as the primary cause of a hypertension.

Viewing the renal lesion separately, aside from the associated cardiac lesion, the one clinical finding characteristic of this lesion is hematuria which is fairly constant. If the bleeding is not macroscopic it is microscopic.

3. *Amyloid kidney.*—An amyloid kidney is a manifestation of general amyloid disease. The amyloid, greater or lesser in amount is deposited almost entirely in the blood vessels, in the glomeruli, the arteriae rectae and the larger vessels. Usually, there is a profound fatty or lipoid degeneration of the tubular cells; occasionally the amyloid is deposited in a kidney the seat of a widespread connective tissue change.

Hypertension, hypertrophy of the left ventricle and retinitis are absent in amyloid disease of the kidneys. As a rule, the urinary findings are fairly characteristic. The urine is light in color and of a low gravity; albumin is plentiful, and casts especially of the waxy variety are fairly abundant. The nephritic signs are usually so overshadowed by the underlying general condition, usually tuberculosis, that they play a small rôle in causing the death of the patient. Such patients die, as a rule, from cachexia.

CONCLUSIONS

1. Clinical findings in nephritis cannot be interpreted in terms of morphologic pathology, except to only a very limited degree. This is due to (a) various extrarenal factors; (b) to the fact that the "type" of the kidney as found at necropsy does not represent an end product but a stage in a fairly well defined pathogenesis. The lesion begins as a glomerulonephritis and all subsequent stages down to the contracted kidney can be traced and are explainable, according to established genetic and morphologic criteria, from this lesion.

2. Thus the conception is set forth that chronic nephritis is essentially a vascular disease, beginning in the capillaries (glomeruli) and extending therefrom to the larger blood vessels. The term "arteriocapillary fibrosis" devised by Sutton and Gull, appears to fit the nature of the malady better than any in current use. In this light chronic nephritis and arteriosclerosis are one and the same lesion. Arteriosclerosis is conceived not as a disease affecting only the larger vessels, but of the entire arterial system from the capillaries to the aorta.

3. The lesions in albuminuric retinitis are analogous morphologically with those of chronic nephritis. This analogy and the sequential relation of albuminuric retinitis to hypertension is a strong argument in favor of the previously submitted thesis that nephritis is not the primary cause of hypertension but is a secondary consequence to the hypertension itself or of the cause or causes (unknown) of the hypertension.

4. The presence or extent of albuminuria bears no consistent relationship to the degree or variety of anatomic destruction of the kidney. Physiologic conditions are more important factors in causing albuminuria than anatomic disturbances in the kidney.

5. The presence or degree of hypertension bears no consistent relationship to the degree or variety of anatomic destruction of the kidney. The probability is very strong that *clinically* cases of chronic nephritis begin as cases of "essential hypertension." If patients with essential hypertension are followed over a long period of time there progressively develop the clinical evidences of either a generalized arteriosclerosis, a localized arteriosclerosis (cerebral endarteritis, coronary sclerosis, albuminuric retinitis) or a renal arteriocapillary fibrosis (chronic nephritis) or a combination of these.

6. The only variety of chronic nephritis that can be reasonably termed an end result is the so-called "contracted" kidney. The contracted kidney arising from a chronic glomerulonephritis (secondary contracted kidney) is morphologically indistinguishable from the "primary contracted" kidney. A "contracted" kidney may be unassociated with hypertension, left ventricular hypertrophy or evidences of renal insufficiency. In such instances, the patient does not die a "renal" death. This kidney is termed the "decrescent" kidney since it corresponds clinically to the "decrescent" variety of arteriosclerosis described by Allbutt.

7. The most important extrarenal factor in modifying renal insufficiency in "arteriocapillary fibrosis" is circulatory insufficiency. Other things being equal, improvement in circulatory insufficiency is coordinate with improvement in renal insufficiency, and vice versa.

8. There is a striking parallelism between the signs of renal insufficiency in arteriocapillary fibrosis associated with hypertension and those noted in frank valvular lesions of the heart. Decompensated phases of valvular heart disease may show evidences of renal insufficiency indistinguishable from those witnessed in chronic nephritis.

9. As a consequence of these studies the following clinical pathologic classification of chronic nephritis is submitted: 1. Arteriocapillary fibrosis. 2. Chronic glomerulonephritis of subacute bacterial endocarditis. 3. Amyloid kidney. 4. "Chronic parenchymatous nephritis" or nephrosis (Epstein).

A CASE OF MASSIVE LIPOMA OF THE MEDIASTINUM

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The presence of a lipoma in the anterior mediastinum, weighing 17 pounds 6 ounces, is so unusual that I wish to record my notes of a case in full.

REPORT OF CASE

R. K. B., aged 37½ years, American; clergyman; married. Family history negative. Aside from diseases of childhood, patient was a healthy child and an athletic boy. At 16 he was kicked in the abdomen by a horse, and his recollection of this was a feeling of distress, at times, for about one year. Never had an injury of the chest; never had pleurisy, pneumonia or pericarditis. He had a deep cough, similar to the one developed with the last illness, about five years before, lasting about a month.

Present Illness.—Patient was well until about seventeen months ago when he sought medical advice for a deep, hollow cough which appeared while in apparent physical fitness. Gradually, this cough became more persistent and was referred to the throat, just above the sternum, described as of an irritating character. Color was good, no constitutional symptoms, no laryngeal symptoms, but an occasional deep sigh and a sensation of a slight substernal oppression. In about four months this amounted to shortness of breath and an occasional wheezing. He slept quietly and well, preferably on the left side, with little or no cough at night.

During the next five months the cough and dyspnea grew rapidly worse and with it a beginning feeling of heaviness in the chest, and exertion was accompanied by an alarming dyspnea. It became necessary to sit upright, or leaning forward. Almost constantly leaning forward; he could not lie on his back. Weight loss, 10 pounds.

At nine months, roentgen-ray treatment being instituted, his condition seemed to improve for a few weeks, but while his cough became less harassing, the dyspnea and oppression again returned and increased. He grew weaker and thinner in the face and arms, slight edema of the lower limbs appeared, and he could sleep only in the knee chest position, while the slightest exertion produced painful attacks of respiratory embarrassment.

By the fifteenth month he had to lean forward resting his chest on a table constantly, the air-hunger was continuous. The edema of the lower extremities, genitalia and abdomen was extreme. Marked cyanosis of the upper extremities and especially the head appeared; later, numbness of the arms, mental torpor, and finally, a distressing struggle for air preceded a mechanical death.

REPORT OF ROENTGENOGRAPHIC EXAMINATION OF THE THORACIC CAVITY

This examination was made at nine months by Dr. Walter C. Barker. His report was as follows:

The anteroposterior view shows a shadow of greater density than normal, extending from the third rib to the diaphragm. This shadow fills the whole breadth of the chest, except a small area of the lower right side. The lateral view shows a dense shadow with a smooth and sharp outline, bulging the anterior wall of the thoracic cavity and extending downward and backward from the third rib to the diaphragm. At the broadest part it almost reaches

posteriorly to the spinal column. The heart shadow is pushed downward and backward, and the lung shadow is mostly above, with a small air space showing below.

Diagnosis: Large primary tumor in the anterior mediastinum.

The record of early physical signs are unobtainable. A persistent hollow cough, increasing in severity and accompanied by an occasional shortness of breath, and later a pronounced dyspnea, constitute the earliest referable symptoms. Yet at nine months, physical examination

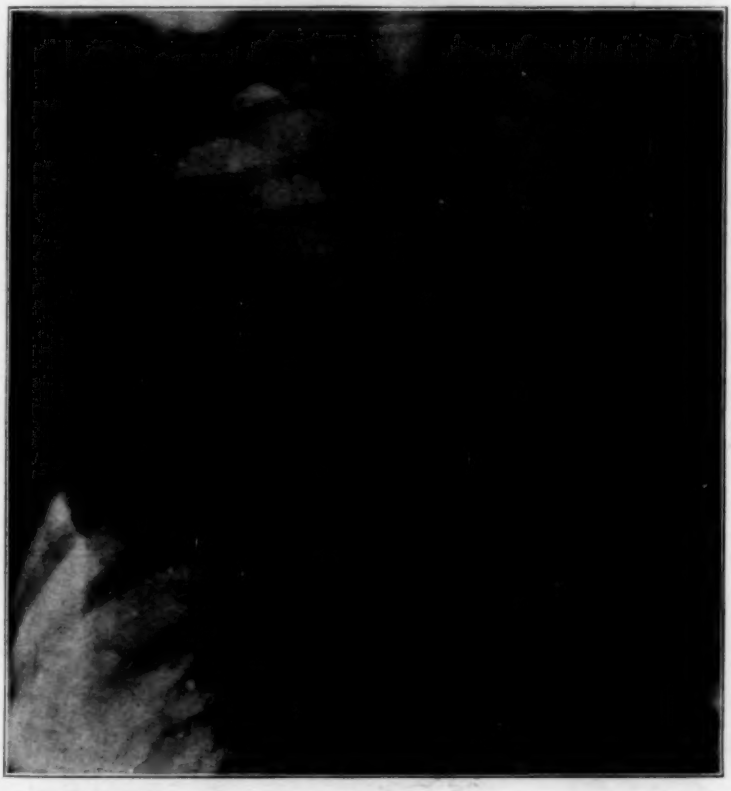

Fig. 1.—Anteroposterior view of chest showing a large tumor. The dark shadow in the lower part of the thorax is cast by the heart; the light shadow at the right and above at the apices is cast by the lungs.

and roentgen-ray examination demonstrated a tumor filling about four-fifths of the chest. There was no pain, no positive emaciation or cachexia, no metastasis, no dysphagia, no aphonia.

Necropsy.—A necropsy was made five and one-half hours after death. Rigor mortis absent. Well developed body, but head and arms thin and of a dusky

bluish hue. Remainder of body extremely edematous. Lower extremities water logged and of twice their normal size. Thoracic cavity: pressure increased, entire cavity apparently filled with an indefinite, lobulated mass conforming to the thoracic arch and extending from the apex to the diaphragm. This mass seems to be attached only at one point, just below the sternal notch, and by only a frail strand of loose adhesivelike bands, easily dissected free with the fingers. The base and posterior surface being found free, the entire

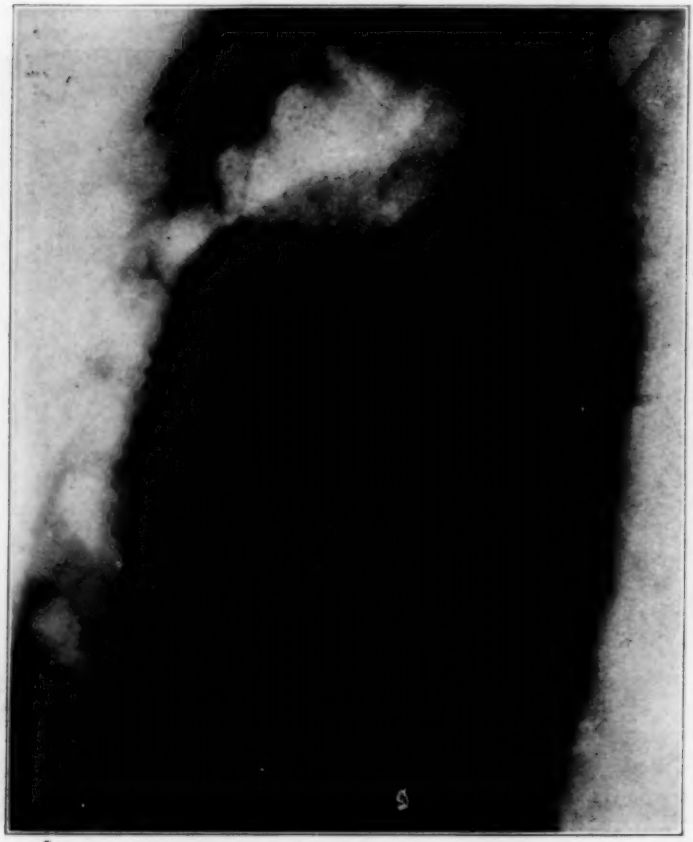

Fig. 2.—Lateral view of chest; shows the shadow of the tumor filling a large part of the thorax. The lungs are crowded above and behind the tumor, and the heart is pushed back and below; the growth being in the anterior mediastinum.

mass is easily delivered, disclosing the heart vessels and lungs compressed along the vertebral column and low in the posterior mediastinum. The heart is very small and pale, measuring approximately 7 by 8 by 6 cm. Lungs measure approximately, 5 by 15 by 3 cm., very dark, and of a liverlike con-

sistence with little crepitation. The peritoneal cavity is filled with a pale transudate, while all the abdominal viscera exhibit extreme degrees of passive congestion.

Pathologic Findings.—A sharply defined, soft fluctuating tumor, covered by a dense tough fibrous capsule which seems to divide it into multiple rounded lobules. On cross section it is apparently a mass of pale yellowish, greasy, fatty tissue. Weight, 17 pounds 6 ounces, measuring 31 by 30 by 15 cm. The anterior surface is convex conforming in general outline and appearance to the thoracic concavity, narrowed at the apex and broad and squatty at the base, with a broad notch midway of the base. The posterior surface is irregular in outline with a vertical concavity about midway, approximately 15 cm. wide at its widest point and about 8 cm. deep. This concavity, which housed the heart, vessels and lungs, is more or less completely lost at the apex while fusing with the notch at the base. All fresh sections stained with sudan III a uniform deep red with few fine grayish stripings of connective bands. The entire tumor developed and fixed well in Kaiserling.

Microscopically, the tumor is composed entirely of masses of fat cells lying in a vascular connective tissue matrix. The individual cells in general are

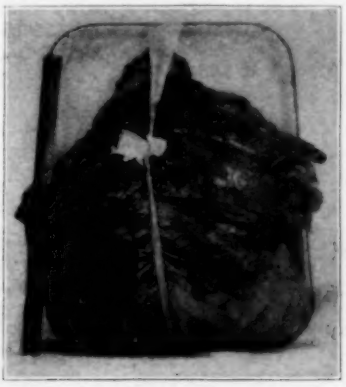

Fig. 3.—The tumor after removal at necropsy, supported against a waiter with a 12-inch ruler standing beside it.

larger than usual fat cells and are round or oval, although in sections taken from about the base they are compressed and flattened. All sections stain perfectly with osmic acid—sudan III.

Medical literature reveals a surprising scarcity of contributions of primary tumors of the mediastinum.[1] While occasional records of teratoma,[2] dermoid,[3] cysts,[4] fibroma and others tumors[5] appear. I have been able to find only four cases of lipoma reported.

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1. Behrens: Weekly Bull. St. Louis M. Soc. **139**: 1911.
2. Atlas: Allg. Wien. med. Ztg., 1894. Becker: Internat. Clin. **19**: 1919.
3. Godlee: Proc. Roy. Med. Soc., London, 1908. Kaestle: München. med. Wchnschr. **1**: 1909.
4. French: Proc. Roy. Med. Soc., Lond., 1907. Blackader: Arch. Pediat. **28**: 1911. Rose: Lancet **2**:1308, 1893.
5. Hare: Mediastinal Disease, Philadelphia, Lea & Febiger, 1889. Edwards: Arch. Pediat. **6**:1889. Roberts: Lancet **2**: 1912. Nichols: Boston M. & S J. 1897.

Beatson [6] records having removed a lipoma of about the size of an orange from the superior mediastinum of a man, aged 45. This presented just above the manubrium sterni and had been increasing slowly for five years, but was not causing any inconvenience. Hare [5] collected three cases, of none of which was he able to obtain a thorough description. All patients were males, the ages of two were given as 38 and 50. In one case the tumor was specified as located in the anterior mediastinum. All patients had marked dyspnea. One patient had marked cyanosis of the face. In two cases the pericardium was involved. The duration of one case is given as a little more than one month. All patients died.

6. Beatson: Glasgow M. J. **1**:57, 1899.

NARCOTIC DRUG ADDICTION

I. THE FORMATION OF PROTECTIVE SUBSTANCES AGAINST MORPHIN *

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The importance that has been attached to the problem of drug addiction in the past several years has prompted us to make a critical study of certain of its phases. While a considerable amount of experimental work has been done on the subject, it has not been wholly satisfactory in that the conclusions have been either conflicting or uncorroborated.

The first phase of the subject we undertook to study was the question as to whether the continuous taking of morphin causes the presence in the blood serum of any substance which has a protective effect against morphin.

LITERATURE

In 1897, Gioffredi,[1] an Italian, published results of experimental work on alkaloidal immunization, in which he stated that while it was impossible to obtain immunity against the effects of cocain and atropin he was able to obtain a serum from a morphin tolerant dog which protected a second dog from almost twice the fatal dose of morphin. Gioffredi[2] published a second paper in 1899, in which he reports further experimental work on the subject. He established a high degree of tolerance in a dog (a tolerance which enabled the animal to withstand three times the minimal fatal dose of morphin), injected the serum of this dog into young cats and determined the degree of protection afforded by it against morphin. His figure for the minimal fatal dose of morphin hydrochlorid in young cats by peritoneal injection is 0.05 gm. per kilogram weight of animal. Ten c.c. of the serum given intraperitoneally protected cats against doses of from 0.05 to 0.07 gm. per kilogram given not more than two hours before the serum, from 0.08 to 0.1 gm. given not more than one hour before the serum, and from 0.11 to 0.13 gm. given from fifteen to thirty minutes before the serum. If the serum was given before the morphin, death did not occur from doses of from 0.05 to 0.14 gm. per kilogram and this he claims to be true even when the serum is administered as long as from ten to fifteen days before the morphin.

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* From the Department of Pharmacology, University and Bellevue Hospital Medical College.

1. Gioffredi: Arch. ital. de biol. **28**:402, 1897.
2. Gioffredi: Arch. ital. de biol. **31**:398, 1899.

Hirschlaff [3] appears to have confirmed to some degree Gioffredi's work. He established a tolerance for morphin in rabbits, and used their serum as a protective substance, employing mice to measure the degree of protection afforded. A study of his tables shows that he considers a dose of from 0.0005 to 0.0007 gm. of morphin per gram weight of mouse as an effective fatal dose. In one series of mice given 1 c.c. of normal rabbit serum, he found no change in his fatal dose. In another series of twenty-one mice treated with 1 c.c. of serum of tolerant rabbits and given doses of morphin varying from 0.0005 to 0.00145 gm. per gram weight, he found that all lived. Some of the serum used to test the protective effect was taken from rabbits just after receiving their last dose of morphin, and some from rabbits which, after acquiring a tolerance, had been deprived of morphin for as much as four weeks before the blood was drawn. There was no difference in the protective effect between the two sera.

Morgenroth,[4] in 1903, pointed out that the dose assumed by Hirschlaff to be a fatal dose is incorrect. Using much greater exactitude in his work he found the fatal dose to be 0.00086 gm. of morphin hydrochlorid per gram of mouse for mice weighing less than 15 gm. In mice over this weight, a dose of 0.00093 gm. per gram of mouse was necessary. He next fixed the fatal dose for mice which twenty-four hours previously received 1 c.c. of normal serum as being 0.0011 gm. per gram of mouse. Finally, after having established the new fatal dose, he tested the protective effect of the serum of rabbits, goats and dogs in which he had established a high degree of tolerance for morphin. From the results on a large series of carefully controlled experiments he definitely concludes that this serum does not possess any specific protective effect.

In 1903 Cloetta [5] brought forth the fact that animals tolerant to morphin lose their tolerance very quickly after withdrawal of the morphin. He states that pigeons and rats accustomed to tolerate 0.15 gm. of morphin without the occurrence of symptoms will succumb to these same doses as soon as two days after the withdrawal. He considers this fact to be in direct conflict with Hirschlaff's findings since one might presuppose that animals whose serum retains the high degree of protection described by Hirschlaff, even four weeks after the withdrawal of the morphin, would be able to preserve their own immunity for at least two days. In order to convince himself whether it were not possible that other substances are formed, direct by-products of morphin, having physiologic antidotal powers, he performed a series of experiments. He poisoned animals with very

3. Hirschlaff: Berl. klin. Wchnschr. **39**:1149, 1177, 1902.
4. Morgenroth: Berl. klin. Wchnschr. **40**:471, 1903.
5. Cloetta: Arch. f. exper. Path. u. Pharmakol. **50**:453, 1903.

large doses of morphin with the hope of obtaining a correspondingly strong antidotal formation, and then used the serum of these animals on mice for the purpose of immunization. Dogs and rabbits were given intravenously large toxic doses of morphin hydrochlorid and after varying periods of from two to seven hours after the last dose, the animals were killed, and the blood drawn and the serum was collected. This serum was then injected into mice in doses of 1 c.c., followed in about twenty-four hours by toxic doses of morphin hydrochlorid. His results show that no such antidotal substance is formed.

Mirto[6] in 1905, in a series of experiments on precipitin reactions using the serum of tolerant rabbits and the various salts of morphin, reaches the conclusion that specific precipitins are not formed.

In 1907, von Marikovszky[7] reported that, after reviewing the work of Hirschlaff and Morgenroth, he thought that a dose of 20 mg. per mouse should be absolutely fatal, but on testing one mouse was astonished to find that the animal survived. From this he assumed that a dose of 30 mg. was the necessary dose. He injected three mice with 30 mg. morphin and followed this with an injection of serum of a rabbit tolerant to morphin (the dose of serum injected is not given). A control mouse received only the 30 mg. of morphin. All the mice died, the only difference being that the serum treated animals survived the control by a few hours. He then experimented with guinea-pigs and found the fatal dose to be 0.07 gm. per 100 gm. weight of animal. In studying his results on the protective effects, we find that the dose of morphin given ranged from 0.07 to 0.116 gm. per 100 gm. and the dose of serum from 1 to 10 c.c. His conclusion as to the protective effect is based on the fact that the serum treated animals survived the controls by periods varying from two to four and one-half hours.

Berri and Belgrano[8] in 1911 claim to have established an agressin and also an anti-aggressin, the latter capable of producing immunizing bodies for cocain and morphin. In their experiments on morphin, they determined the minimum fatal dose of morphin hydrochlorid for rabbits to be 0.41 gm. per kilogram by intraperitoneal injection. Their aggressin was prepared by injecting into the pleural cavities of a rabbit an emulsion of aleuronat followed in about ten hours by morphin hydrochlorid in the dose of 0.20 gm. per kilogram. After from twelve to fourteen hours they aspirated the exudate said to contain the aggressin. From this point they subdivided their work into four series of experiments. In the first series they injected subcutaneously three rabbits with 5 c.c. of the exudate, followed in fifteen minutes by

6. Mirto: Arch. farma. sper. **4**:406, 1905.
7. Von Marikovsky: Zentralbl. f. Bakteriol. u. Parasitenk. **43**:494, 1907.
8. Berri and Belgrano: Annali dell Instituto Maragliano **5**:42, 1911.

0.36 gm. morphin hydrochlorid per kilogram intraperitoneally. The animals all died within a period of three and one-half hours. As controls they injected three rabbits with 0.39, 0.41 and 0.415 gm. per kilogram doses of morphin (without the exudate), respectively. The results were that the first animal lived while the other two died in fourteen and ten hours, respectively. Another animal received 7 c.c. of the exudate alone, and remained alive without presenting any signs of poisoning. From this they conclude that they have obtained an aggressin.

In the second series they studied the immunizing properties of the aggressin itself. They prepared three rabbits by giving each five subcutaneous injections of the aggressin in increasing doses from 3 to 6 c.c. at intervals of from four to five days. Five days after the last dose of aggressin, an intraperitoneal injection of morphin was given, the doses being 0.415, 0.42 and 0.43 gm. per kilogram, respectively. All the rabbits showed toxic effects of the morphin, the first two recovering while the one receiving 0.43 gm. per kilogram died in forty-eight hours. A control rabbit received 0.41 gm. per kilogram of morphin and died in fifteen hours.

The third series shows a study of the protective effect of the serum of rabbits immunized with five injections of increasing doses of the aggressin as in the preceding experiment. Five days after the last dose the animals were bled and the serum collected. Three rabbits were given subcutaneously 10 c.c. of this serum followed in twenty-four hours by 0.415 gm. per kilogram morphin in one of the rabbits and by 0.42 gm. per kilogram in the remaining two rabbits. The first rabbit lived indefinitely, the remaining two died in five and seven days, respectively. A control receiving 0.41 gm. per kilogram morphin died in fifteen hours.

The last series of experiments were performed to show the production of anti-aggressin substances in rabbits. Three rabbits were injected subcutaneously with serum of rabbits immunized against aggressins, two receiving 10 c.c., and one 60 c.c. After twenty-four hours, a subcutaneous injection of 5 c.c. of aggressin was made and followed in fifteen minutes by intraperitoneal injection of morphin in doses of 0.36, 0.37 and 0.37 gm. per kilogram, respectively. All showed toxic effects but recovered. A control receiving 5 c.c. of agressin plus 0.36 gm. per kilogram of morphin died in three hours.

Von Egmond [9] in 1911 and also Van Dongen [10] in 1915 show that morphin tolerance is partially selective in character. Certain parts of the central nervous system (the vagus center) and the gastro-

9. Von Egmond: Arch. f. exper. Path. u. Pharmakol. **65**:197, 1911.
10. Van Dongen: Arch. f. d. ges. Physiol. **162**:54, 1915.

intestinal tract do not show the same degree of tolerance as do the brain cortex, the vomiting center or the respiratory center. In fact, in dogs tolerant to 1 gm. of morphin it was possible to change the heart rate from 120 to 80 per minute by a dose of 0.04 gm. Doses as small as 0.00004 gm. per kilogram in these same animals showed an appreciable effect.

EXPERIMENTAL WORK

In our work we made use of serum from two sources, first from humans addicted to morphin, and secondly from dogs which were made tolerant to morphin. In the case of the human serum, the blood was drawn while the person was still addicted, and as long as possible after the last dose of morphin. In either case the blood was drawn, allowed to clot and the serum separated by centrifugalization. The serum was injected into the animals as soon as possible after the drawing of the blood.

In setting forth these experiments we subdivide them, for convenience, into two large groups: (1) the effect on mice, and (2) the effect on cats. We chose mice and cats especially as the most suitable animals on which to determine the protective effect of the serum because most of the previous work had been done on these animals.

Group 1: *Mice.*—Our first attempt was to repeat some of the work appearing in the literature, but after a number of trial tests on mice we found it essential to determine to our own satisfaction the minimal fatal dose of morphin sulphate per gram weight of mouse. Anyone who has worked on the determination of a definite minimal fatal dose of a drug must recognize the inherent difficulties of the problem. Differences in the susceptibility of individual animals, errors in technic, differences in the rate of absorption of the injected poison, render it impossible to obtain a figure which truly represents the actual minimal fatal dose, that is the smallest dose which will invariably cause death.

These difficulties became so apparent in the course of our work, some mice surviving a dose of 1 mg. per gram, while others succumbed to 0.2 mg. per gram, that we finally decided to ascertain the dose that would kill a majority of mice, and base our conclusions on a comparison of the percentage of fatalities from such a dose with and without the serum. The words "minimal fatal dose," used hereafter, mean such a dose.

In performing the tests for the minimal fatal dose in mice, we adhered strictly to the following points in technic.

1. The mice were deprived of food and water for a period of about eighteen hours before weighing and injecting. This was to obviate any error in dosage which might be caused by the weight of the contents of the stomach, intestine and bladder. The loss in this period

of time averaged from 2 to 3 gm. per mouse. The deprivation of fluids also allowed a rapid and fairly uniform rate of absorption of the fluid injected.

2. The injection was made into the subcutaneous tissues of the abdomen, and the skin at the site of the puncture was pinched while the needle was being withdrawn and was so held for at least one-half minute after the withdrawal of the needle. This was done to prevent any of the injected morphin solution from oozing out, as we found that the loss of one drop was sufficient to change the results.

3. The amount of morphin sulphate solution to be injected was made up to 0.4 c.c. This was accomplished by making a 4 per cent. solution of morphin sulphate and diluting the amount of solution required with distilled water to 0.4 c.c. Furthermore, the 0.4 c.c. of morphin solution was injected into two different places, 0.2 c.c. in each, the needle being partly withdrawn after the injection of the first 0.2 c.c. and then reinserted at a different angle for the second injection of 0.2 c.c. We found this method to give us the most uniform rate of absorption. In our early trials we made use of a straight 4 per cent. morphin sulphate solution, making a single injection with the idea that the amount of fluid injected should be in direct proportion to the body weight of the mouse, thereby eliminating the factor of dilution of the blood. However, we soon discovered that the factor of dilution was less important than the factor of uniformity of absorption. In injecting a like total amount of fluid in each mouse regardless of weight, the tension of the confined fluid injected and the area exposed for absorption are the same for every one of the mice of a series. These two factors have a marked bearing on the uniformity of absorption.

4. In order to gain greater accuracy we used a standardized 1 c.c. tuberculin syringe and needles one and one-half inches long, which were inserted their full length before injecting. The morphin sulphate solution was freshly prepared for each experiment. It might be interesting to note in this connection that in determining the depression of the freezing point of a 4 per cent. solution of morphin sulphate, we found it to be hypotonic as compared to a physiologic sodium chlorid solution.

It is unnecessary to tabulate all our results, only those which approximate our minimal fatal dose. Table 1 shows the results of the minimal fatal dose tests.

We took as our end reaction, as to the time of death, only the mice which died within 4 hours. The majority of mice died within this time; the ones surviving had usually recovered from the acute stage of poisoning in so much as they were able to take food. Very

few of the mice in our series died after the four hour period. This we ascribe to causes other than the acute intoxication. Table 2 shows the time of death of the mice enumerated in Table 1.

These two tables show that of animals receiving 0.3 mg. or less of morphin sulphate per gram weight, the majority die. In other words, the minimal fatal dose of morphin sulphate as closely as we can determine it is between 0.3 and 0.4 mg. per gram weight of mouse. Any protective effect of the serum ought, therefore, to be shown definitely with a dose at or about 4 mg. per gram. The proportion of mice surviving this dose should be noticeably greater with the serum than without, if the latter has any immunizing properties whatever.

TABLE 1.—Results of Minimal Fatal Dose Tests

Dose of Morphin SO_4 per Gm. Weight	No. of Mice Tested	No. Mice Dead Within 4 Hours	Per Cent. of Deaths
0.2	20	7	35
0.3	16	5	31
0.4	30	18	60
0.6	10	8	80

TABLE 2.—Time of Death

Time of Death in Hours	Dose of Morphin Sulphate per Gm. Weight of Mouse				
	0.2	0.3	0.4	0.6	Total
0- 1	0	2	0	0	2
1- 2	3	1	10	2	16
2- 3	4	2	7	6	19
3- 4	0	0	1	0	1
4-24	2	0	1	2	5

Having determined the smallest dose of morphin sulphate which is fatal to the majority of mice receiving it, we proceeded to a determination of the protective action of the serum.

In one series of experiments we made use of the serum of a morphin tolerant dog. This animal maintained an approximate weight of 5 kilograms throughout the period of morphinization. He was started on a dose of 10 mg. daily and this dose was gradually increased until at the end of three and one-half months the animal was receiving 80 mg. daily. In the beginning, the 10 mg. produced definite symptoms; at the end of this period the 80 mg. produced none. This final dosage reached in the animal would correspond approximately to a gram per day in the average human which represents the usual amount taken by morphin addicts. When the blood was drawn the dog had received no morphin for a period of eighteen hours.

The technic followed with the mice is the same as described above, with the exception that after weighing the mice, 1 c.c. of the serum was injected into the subcutaneous tissues over the back, and after a period of from three to four hours the required amount of morphin sulphate was injected.

TABLE 3.—Results of Injection of Serum and of Morphin

Mouse	Weight of Mouse in Gm.	Time of Injection of Dog's Serum	Time of Injection of Morphin Sulphate 0.4 Mg. per Gm.	Result
1	25.5	12: 20	4: 04	Died 6: 45
2	26	12: 19	4: 02	Survived
3	20	12: 19½	4: 03	Died 6: 15
4	25.5	12: 20½	3: 58½	Died 6: 10
5	25.5	12: 21	3: 59½	Survived
6	21.5	12: 22½	3: 43	Died 6: 20
7	21.5	12: 22	3: 41½	Died 5: 35
8	21.5	12: 28	3: 44½	Survived
9	21.5	12: 27½	3: 47	Died 6: 00
10	21.5	12: 27	3: 45½	Died 6: 00

This table shows that serum of the morphin tolerant dog did not convey any immunity to the mice.

The analysis of the figures of this series show that 70 per cent. of the mice died within the four hour period, which is slightly more than in the control experiment of mice receiving no serum as shown in Table 1. One must conclude, therefore, that the serum of a morphin tolerant dog does not contain any protective substance against morphin.

In a second series we duplicated the experiments of Series 1, using the serum of a human addict, who had been taking morphin for six years and whose daily doses ranged from 10 to 12 grains of morphin sulphate. A period of three and one-fourth hours elapsed between the last dose of 2½ grains and the drawing of the blood. The patient at this time was very uncomfortable and felt that he was in great need of his morphin.

The serum from this blood was injected into a series of mice in 1 c.c. doses between 5:00 and 5:20 p. m.; the required amount of morphin sulphate was injected the next morning as shown in Table 4. The technic was the same as above stated with the exception as to the time of injection of the serum.

Here again we find a mortality of 70 per cent on a dosage of 0.4 mg. morphin sulphate per gram weight, following an injection of a human addict's serum. These results show that the serum had no protective substance against the morphin. Similar results were obtained with doses of 0.3 mg. of morphin sulphate with the same serum as used above; out of six mice injected, three mice dying and three surviving.

Group 2: Cats.—In this series of experiments we simply attempted to repeat the work of Gioffredi on cats, using, however, the serum of human addicts who were under strict hospital supervision; the blood being drawn as long as possible after the last dose of morphin.

Our technic was as follows: The cats were first weighed, the serum injected into the external jugular vein without any anesthetic, and at varying periods of time the required dose of morphin sulphate was injected subcutaneously.

TABLE 4.—Result of Injection of Morphin Following Injection of Serum

Mouse No.	Weight of Mouse in Gm.	Time of Injection 0.4 Gm. Morphin Sulphate	Result
1	24	11: 27½ a.m.	Died 1: 48 p.m.
2	24	11: 29½ a.m.	Died 1: 33 p.m.
3	23	11: 06 a.m.	Died 1: 35 p.m.
4	22	11: 10 a.m.	Died 12: 55 p.m.
5	20	11: 23 a.m.	Died 12: 35 p.m.
6	20.5	11: 21 a.m.	Died 1: 30 p.m.
7	20	11: 37 a.m.	Died 2: 05 p.m.
8	20	11: 33 a.m.	Survived
9	19.5	11: 41 a.m.	Survived
10	22	11: 16 a.m.	Survived

We divided the time for the injection of the serum into four different periods taking the time of the injection of morphin as our starting point: (1) 30 minutes after the morphin injection, (2) simultaneously with the morphin injection; (3) two hours before the morphin injection, and (4) from eighteen to twenty-four hours before the morphin injection. In determining the protective effect of the serum, we took the figures 0.07 and 0.08 gm. of morphin sulphate per kilogram weight of animal given subcutaneously, as the dose which should be survived if the animal were protected by the serum. We used these doses inasmuch as Geoffredi claims that he was able to protect young cats from doses varying from 0.05 to 0.14 gm. of morphin per kilogram given intraperitoneally.

We did not attempt to determine the minimal fatal dose for cats on the same principle as that followed with the mice, because of the difficulty of getting the larger number of cats required, and because Gioffredi's results could be more directly tested by taking a sure fatal dose well within his figures.

Tables 5, 6, 7 and 8 give the results of our experiments.

TABLE 5.—Effect of Morphin Sulphate Given Before the Serum

Cat	Weight in Kg.	Dose of Morphin Sulphate per Kg. Gm.	Time of Injection of Morphin Before Serum	Amount Serum	Death
5	2.47	0.08	22 minutes	10 c.c.	2½ hrs.
6	2.23	0.08	33 minutes	10 c.c.	2 hrs.

TABLE 6.—Effect of Morphin Sulphate Mixed with the Serum and Given Subcutaneously *

Cat	Weight in kg.	Dose of Morphin Sulphate per Kg.	Dose of Serum	Death
7	3.0	0.08	10 c.c.	4 hours
8	2.7	0.08	10 c.c.	Survived

* In this experiment we allowed the mixture of morphin sulphate and serum to stand over night before injecting. In the morning we found that the morphin had precipitated out. On mixing morphin sulphate solution with normal human serum in the same proportions, we found that precipitation also occurred. We therefore do not feel that this is a specific effect of addict's serum but due, most likely, to the alkalinity of the blood, or its salts reacing with the morphin.

The survival of cat 8 may be attributed, we think, to the fact that absorption was much delayed by the precipitation of a good part of the morphin salt.

TABLE 7.—Effect of Morphin Sulphate Given a Short Period After the Serum *

Cat	Weight in Kg.	Time Serum Given Before Morphin	Dose Serum	Dose Morphin Sulphate per Kg.	Death
9	3.06	2 hours	10 c.c.	0.07	Over night
10	3.0	1 hour	10 c.c.	0.07	Over night
11	2.43	1 hour	8 c.c.	0.07	2½ hours

* In the cats that died over night, we do not know the exact time of death, as they were injected very late in the afternoon and no observations were made until our return to the laboratory the next morning. In both cases the cats had been dead for a considerable period of time as they were cold and very rigid.

TABLE 8.—Effect of Morphin Given from Eighteen to Twenty-Four Hours After the Serum

Cat	Weight in Kg.	Dose of Serum	Dose Morphin Sulphate per Kg.	Death
2	1.97	10 c.c.	0.08	2 hours
3	1.52	10 c.c.	0.08	4 hours
4	2.13	12 c.c.	0.08	3 hours
12	3.37	10 c.c.	0.07	2½ hours
13	2.76	9 c.c.	0.07	2½ hours
14	2.73	10 c.c.	0.07	1½ hours

SUMMARY

We wish to emphasize the following features of our work:

1. The difficulty attendant in determining a minimal fatal dose of a drug, especially calling attention to the factor of uniformity of absorption of subcutaneous injections.

2. That the minimal fatal dose of morphin sulphate for mice is a question which depends very largely on the technic followed; with our technic it proved to be between 0.3 to 0.4 mg. per gram weight of mouse.

3. The acute stage of morphin intoxication has reached its height within four hours after the subcutaneous injection, and recovery usually occurs if this period is survived.

4. That serum of the blood of a dog tolerant to morphin sulphate does not protect mice against a minimal fatal dose of morphin sulphate.

5. That the serum of the blood of human addicts does not protect mice against a minimal fatal dose of morphin sulphate.

6. That 10 c.c. of serum of humans addicted to morphin did not protect cats from doses of from 0.07 to 0.08 gm. of morphin sulphate per kilogram regardless of the sequence or of the interval of time elapsing between the injection of the serum in relationship to the injection of the morphin sulphate.

7. The morphin tolerance does not develop immunizing properties in the serum either of dogs or of human beings.

DISCUSSION

It will be seen that our work gives results which are in direct conflict with the claims of Gioffredi, Hirschlaff and von Marikovsky, all of whom believed that they had established definite protective substances in the serum of morphin tolerant animals.

In studying the work of Gioffredi, especially as reported in his second paper, we find an absence of tables and protocols, making it impossible to subject his experiments to a critical analysis. His results as stated by him appear convincing, but are not presented in such a form as to permit of intelligent discussions, and amount to little more than statements of conclusions. It may be noted that no attempt has been made in that long period since the publication of his report, to confirm his work by experiments on cats.

Our own experiments (Tables 5, 6, 7 and 8) permit of direct comparison with his stated results, with the difference that he used the serum of a single tolerant dog, whereas we used the serum of a number of tolerant human beings. Except for the possibility that the serum of the dog used by him happened to possess some peculiar quality, not due to the tolerance, it is impossible to reconcile his results with ours.

He states that if the serum is injected within one hour after the morphin it will protect against doses ranging from 0.08 to 0.10 gm. per kilogram; and if within from fifteen minutes to half an hour, against doses ranging from 0.11 to 0.13 gm. per kilogram. If injected simultaneously it would protect against doses up to 0.13 gm. per kilogram.

In the experiments analyzed in tables 5 and 6 we used the lowest of all the doses mentioned, and did not find any protective effect, unless the survival of the one cat mentioned in Table 6 be attributed to causes other than precipitation.

Gioffredi further states that if the serum is given before the morphin, it will protect against doses ranging as high as 0.14 gm. per kilogram, and this even if given as long as from ten to fifteen days before.

Tables 7 and 8 show no protection against doses as small as 0.07 and 0.08 gm. when the serum was given from one hour to twenty-four hours before the morphin.

The work of Hirschlaff does not require extended comment, as Morgenroth, using the same principle, viz., the determination of a *surely* fatal dose, and testing the protective power of serum against such a dose—found that Hirschlaff's results were due principally to poor methods of determining the fatal dose. We feel that Morgenroth's work clearly proves that no credit is to be attached to Hirschlaff's conclusions, but our own preliminary experiments made us doubt the validity of any conclusions to be drawn, either way, on the basis of the principle of determination of the fatal dose employed by both these workers.

We found that the range of individual suceptibility to morphin in mice is so great that a dose surely fatal to all mice would probably be many times the amount required to kill the average mouse, and we therefore, as set forth above, adopted a different principle, viz., the comparison of percentages of fatalities with and without serum under a given dose previously ascertained to be fatal to a majority of mice. This we believe to be a much more delicate method, and one from which reliable conclusions can be drawn.

Our comment on the work of Hirschlaff and Morgenroth may be summarized by saying that the conclusions of the latter are fully sustained by our own work, following what we believe to be a more reliable method, and that Hirschlaff's conclusions are contradicted by the more careful work of Morgenroth as well as by our own.

The work of von Marikovsky on mice was confined to three mice and one control, and hardly permits of any conclusions being drawn. Von Marikovsky also worked with guinea-pigs, but with these animals, as well as with the mice, his conclusions are based merely on the difference of a short period in the time of death between the serum treated animals and the controls. Our own tests with mice have shown that while a few animals die (apparently not from acute morphin poisoning) after the lapse of four hours, the great majority

either die within that time or definitely recover and survive. We, therefore, do not think that valid conclusions can be drawn from slight differences in the time required to kill.

Berri and Belgrano worked entirely with rabbits, and we are not at present in a position to discuss their work with reference to our own. It may be noted, however, that their method of obtaining the serum claimed to have protective effects was by the production of a substance which they classed as an aggressin, and it is doubtful, in view of what is now understood relative to the so-called aggressins, whether their conclusions could be considered tenable.

The researches of other workers who have attacked the problem in different ways may be referred to as having decided significance. Cloetta's experiments, which were carefully performed, show that the immunity conferred by acquired tolerance is lost very rapidly by the animal itself, and this accords with general experience among human addicts, who show marked symptoms of acute poisoning on attempting to take their previously accustomed dose a few days after withdrawal. If the active immunity, as it may be called by analogy, of the tolerant animal, retaining its entire blood volume, disappears within two days, it is scarcely conceivable that passive immunity could be transferred by a comparatively small injection of serum, and would last, as claimed by Gioffredi, for ten or fifteen days, or as claimed by Hirschaff, four weeks.

Again, as demonstrated by the work of von Egmond and by that of Van Dongen, the sensitiveness to morphin of certain centers of the brain persists after the acquisition of a general tolerance, a fact which is not consistent with the possibility of neutralizing substances in the blood serum.

The researches in immunology bearing on the nature and mechanism of serum immunity have in recent years tended to make it constantly more probable that such immunity is produced only by proteins, and that the tolerance established to certain vegetable alkaloids is of a totally different nature.

CONCLUSION

1. We consider that the definite conclusion to be drawn from our work, which, so far as we know, is the only work of this nature directly testing the question of immunity acquired by the human morphin addict, is that no substance is formed in the blood serum of a human being who has acquired a high tolerance to morphin, which is capable of conferring any degree of immunity to the toxic action of morphin on an animal into which it is injected.

2. Likewise we have been able to show that the blood of a tolerant animal does not contain any protective substance against morphin.

An extensive bibliography on the subject of morphin addiction is contained in an article by Du Metz, J. A. M. A. 72:1069 (April 12) 1919.

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THE MARGIN OF SAFETY OF INTRAVENOUS DIGITALIS IN CATS*

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The medical profession has become greatly interested in intravenous therapy in recent years, and although no particular evidence of this enthusiasm has been reflected in the treatment of heart disorders, the work of Eggleston,[1] introducing the administration of large single doses of digitalis by mouth, might well lead others to try the effect of similar preparations intravenously. In the past, intravenous cardiac therapy has been essentially limited to preparations of strophanthin, which is generally considered to be a dangerous procedure. With this in mind, an attempt was made to determine whether the introduction into the blood stream of digitalis in its more complete and less potent form would prove to be a safe method for obtaining a therapeutic effect.

The margin of safety has been taken as the difference between the minimum lethal dose (M.L.D.) and the minimum toxic dose (M.T.D.). We introduce the concept of the margin of safety of digitalis preparations because in the practical use of the drug the therapeutic dose is very close to the toxic dose. Therefore, it is of great importance to know how far removed the lethal dose is from the toxic dose, and whether the margin is greater in some preparations than in others. The minimum lethal dose is that dose which is just required to cause standstill of the heart and death when the digitalis preparation is given in interrupted doses over a period of about one hour, similar to the method of Hatcher.[2] In a previous work by one of us [3] it was shown that the toxic and lethal doses are independent of the speed of administration of the drug from a period of fifteen minutes to four hours; and, therefore, in this work the injections were not given at any constant speed, sometimes producing the lethal effect in about one-half hour and sometimes in two hours or more. Some difference of opinion might exist as to the occurrence of the toxic effect. It is generally thought that the appearance of heart block, changing ventricular complexes, and extrasystoles indicate toxicity. The latter is the most common event in cats, and this was generally considered in timing the

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1. Eggleston, C.: Digitalis Dosage, Arch. Int. Med. **16**:1 (July) 1915.
2. Hatcher, R. A., and Brody, J. G.: A Biological Standardization of Drugs, Am. J. Pharmacy **82**:360, 1910.
3. Levine, S. A.: The Action of Strophanthin on the Living Cat's Heart, J. Exper. M. **29**:485, 1919.

toxic dose. This criterion has the added advantage that it is the most important clinical sign of toxicity, for heart block in itself rarely impairs the efficiency of the circulation, while extrasystoles indicate an increased irritability of the ventricles, and when numerous, certainly diminish the effective output of the heart. The minimum toxic dose, therefore, is calculated in these experiments as the smallest dose that is required to produce extraventricular systoles.

METHOD OF PROCEDURE

Cats were used in preference to other laboratory animals because their reaction to digitalis is more like that of the human subject. The cat was etherized, placed on the animal board and the right front leg and left hind leg connected with the electrocardiograph. A canula was inserted in the right saphenous vein and attached by a small rubber tube to a 10 c.c. calibrated pipet which served as a buret. The normal electrocardiogram was taken, and the dose of the preparation to be used was then allowed to flow into the vein. One minute before each dose an electrocardiogram was taken, until a toxic effect was expected, previous experience having shown about where this was to occur. During the two critical periods, the beginning of toxic and lethal effects, the movements of the galvanometer string were watched carefully, and frequent tracings were taken to detect the earliest abnormality.

Various different digitalis preparations were used. The first was an aqueous extract of powdered digitalis (Marion, Virginia leaf) prepared according to Pratt;[4] the second was Squibb's powdered digitalis leaves prepared in the same way; the third was Squibb's tincture of digitalis, and finally, three different digitalis preparations in ampoules for subcutaneous and intramuscular use were tested. The details of the experiments are given in the accompanying table. In three experiments the average margin of safety (M.L.D. — M.T.D.) of intravenous aqueous extract of digitalis (Virginia leaf) was 50 per cent., that is, it required approximately half as much of the drug to produce extraventricular systoles as it did to cause standstill of the heart. An identical figure (50 per cent.) was obtained in the average of three experiments using the aqueous extract of Squibb's digitalis leaves. The average for Squibb's tincture of digitalis (five experiments) was 40 per cent.; for digifolin ampoules (one experiment) 50 per cent.; for digalen ampoules (one experiment) 56 per cent., and for digipuratum ampoules (one experiment) 64 per cent. It is evident that there are individual variations in different cats, both in the suscepti-

4. West, H. F., and Pratt, J. P.: Clinical Experience with a Standardized Aqueous Extract of Digitalis, J. A. M. A. **74**:1389 (May 15) 1920.

THE MINIMUM LETHAL DOSE AND MAXIMUM TOXIC DOSE OF VARIOUS DIGITALIS PREPARATIONS

Cat Number	Weight in Kilos.	Drug	Speed of Administration	Toxic (M. T. D.) Dose	Lethal (M. L. D.) Dose	Per Cent. of M. L. D. that is Toxic	M. T. D. per Kilo.	M. L. D. per Kilo.	Margin of Safety M. L. D. minus M. T. D., per Cent.	M. L. D. per Kilo. in Gm. of Digitalis Leaves	Time for M. L. D.
1	2.2	Aqueous extract of Virginia leaf	0.5 c.c. every 6 min.[1]	0.033 gm.	0.06 gm.	55	0.015 gm.	0.027 gm.	45	0.11	1 hr. 56 min.
2	2.4	Aqueous extract of Virginia leaf	1.0 c.c. every 6 min.	0.035 gm.	0.07 gm.	50	0.015 gm.	0.029 gm.	50	0.12	1 hr. 30 min.
3	3.2	Aqueous extract of Virginia leaf	1.0 c.c. every 6 min.	0.030 gm.	0.065 gm.	46	0.009 gm.	0.020 gm.	54	0.08	1 hr. 14 min.
4	4.2	Aqueous extract of Squibb's leaf	1.5 c.c. every 24 min.[2]	0.113 gm.	0.188 gm.	60	0.027 gm.	0.045 gm.	40	0.11	1 hr. 44 min.
5	2.9	Aqueous extract of Squibb's leaf	0.3 c.c. every 6 min.	0.038 gm.	0.105 gm.	36	0.013 gm.	0.036 gm.	64	0.09	1 hr. 10 min.
6	2.6	Aqueous extract of Squibb's leaf	0.3 c.c. every 6 min.	0.06 gm.	0.113 gm.	53	0.023 gm.	0.042 gm.	47	0.11	1 hr. 25 min.
7	2.1	Squibb's tincture digitalis	0.2 c.c. every 12 min.	0.1 gm. (1 c.c.)	0.22 gm. (2.2 c.c.)	45	0.048 gm. (0.48 c.c.)	0.105 gm. (1.05 c.c.)	55	0.105	2 hr. 1 min.
8	3.2	Squibb's tincture digitalis	0.4 c.c. every 6 min.	0.16 gm. (1.6 c.c.)	0.28 gm. (2.8 c.c.)	58	0.050 gm. (0.5 c.c.)	0.088 gm. (0.88 c.c.)	42	0.088	41 min.
9	3.5	Squibb's tincture digitalis	0.2 c.c. every 6 min.	0.2 gm. (2.0 c.c.)	0.34 gm. (3.4 c.c.)	59	0.057 gm. (0.57 c.c.)	0.097 gm. (0.97 c.c.)	41	0.097	1 hr. 44 min.
10	3.4	Squibb's tincture digitalis	0.35 c.c. every 6 min.	0.28 gm. (2.8 c.c.)	0.385 gm. (3.85 c.c.)	73	0.082 gm. (0.82 c.c.)	0.11 gm. (1.1 c.c.)	27	0.11	1 hr. 3 min.
11	3.2	Squibb's tincture digitalis	0.5 c.c. every 24 min.	0.30 gm. (3.0 c.c.)	0.45 gm. (4.5 c.c.)	67	0.094 gm. (0.94 c.c.)	0.14 gm. (1.4 c.c.)	33	0.14	3 hr. 30 min.
12	2.3	Digifoline ampoules[3]	0.25 c.c. every 6 min.	0.175 gm. (1.75 c.c.)	0.35 gm. (3.5 c.c.)	50	0.076 gm. (0.76 c.c.)	0.152 gm. (1.52 c.c.)	50	....	1 hr. 19 min.
13	3.1	Digalen ampoules[4]	0.25 c.c. every 6 min.	0.83 mg.	1.88 mg.	44	0.27 mg.	0.61 mg.	56	....	2 hr. 30 min.
14	1.3	Digiparatum ampoules[5]	0.1 c.c. every 6 min.	0.05 gm. (0.5 c.c.)	0.14 gm. (1.4 c.c.)	36	0.038 gm.	0.11 gm.	64	....	

1. 100 gm. of Virginia leaf gave 24.5 gm. of aqueous extract. 1 c.c. = 0.005 gm. aqueous extract.
2. 100 gm. of Squibb's powdered leaf gave 39.5 gm. of aqueous extract. 1 c.c. of solution = 0.025 gm. aqueous extract.
3. 1 c.c. = 0.1 gm. digitalis leaves; Society of Chemical Industry, Basle, Switzerland. (Ciba Co., New York).
4. 1 c.c. = 0.3 mg. amorphous digitoxin, Hoffmann, La Roche Chemical Works, N. Y. (Made in Switzerland).
5. 1 c.c. = 0.1 gm. of digipuratum powder, manufactured by Knoll & Co., Ludwigshafen. (Merck and Co., N. Y.).

bility to the drug and in the margin of safety, which varied from 27 to 73 per cent. Similar variations are found in the intravenous use of ouabain.[2] Undoubtedly, similar individual variations in susceptibility to digitalis are present in the human and should always be taken into account in practical therapy. The average margin of safety of all the experiments in this study was 48 per cent. This is identical with the result obtained with crystalline strophanthin or ouabain.[3]

The practical consideration that follows from these experiments is that although the various digitalis bodies, when given by mouth, are generally regarded as much safer than intravenous administration of strophanthin, when the entire digitalis glucosids (either the aqueous or alcoholic extracts) are given intravenously, the same risk is encountered as in using strophanthin. In fact, the rapidity with which the drugs act on the heart does not seem to differ very much no matter what preparation is put directly into the circulation. In one experiment, a single lethal dose was given and the toxic effect occurred in two minutes, and standstill in sixteen minutes. Large single doses of strophanthin produce death in from nine to thirty minutes.[3] In two experiments the interval between injections was purposely prolonged to twenty-four minutes to allow the opportunity of determining how soon after an injection the effect appears. It was found that the toxic effect occurred within three minutes after the injection. In experimenting with crystalline strophanthin, an interval of six minutes between injections was found sufficiently long to bring out any changes, that is, waiting from twelve to twenty-four minutes did not alter the results. Practically the same findings were observed using digitalis; i. e., if an effect is to occur at all with interrupted injections it will result within six minutes after the last one.

CONCLUSIONS

1. The average margin of safety (the difference between the minimum lethal dose and the minimum toxic dose) of various digitalis preparations when given intravenously to cats was found to be 48 per cent.

2. The rapidity with which intravenous digitalis acts is similar to strophanthin, and does not differ appreciably no matter what preparation is put into the blood stream.

3. The risk in intravenous digitalis therapy appears from these experiments to be as great as in intravenous strophanthin.

We wish to express our appreciation to Dr. Philip Marvel, Jr., of Atlantic City, N. J., for his kind assistance in the first three experiments.

CONCERNING THE DIAGNOSIS AND TREATMENT OF HYPOTHYROIDISM *

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The results of a comparative study of the clinical and laboratory data for determining thyroid deficiency are reported herewith. There is very little difficulty in recognizing the average well-developed case of myxedema or cretinism, but our observations show that cases of mild thyroid deficiency are probably more numerous than has previously been supposed; also that some will remain unrecognized unless special methods are used in diagnosis and control of treatment.

A series of cases of thyroid disease were selected for study. Certain of these cases had been previously diagnosed definitely hypothyroid. Other patients had long been seeking relief from indefinite ill-health previously undiagnosed, which no form of treatment seemed to alleviate. With the exception of one patient, (Case 1940), a child, 2 years old and too young for special methods of examination, the group could definitely be diagnosed by aid of combined laboratory and clinical methods as thyroid deficiency cases. Four individuals in whom symptoms of thyroid deficiency with thyrotoxicosis occurred were regarded as having thyroid dystrophy and were included in these observations.

METHODS OF PROCEDURE

The usual clinical history was written; physical examination, differential blood count, glucose tolerance test, and basal metabolism determinations were made. A study of nitrogen balance was made in two cases, children too young for basal metabolism determinations with the means at hand. The technic of the glucose tolerance tests was that previously developed by one of us,[1] consisting of the ingestion of 1.5 gm. glucose per kilogram body weight in 3 c.c. water per gram of sugar, a sample of blood being withdrawn immediately before the glucose had been taken, and at intervals of one hour, for two or more hours thereafter. The blood sugar was determined by the method of Folin.[2] The basal metabolism determinations were made with the Benedict portable respiration apparatus, following the technic of Francis G. Benedict,[3] in which all patients are required to come to the

* From the Memorial Metabolism Clinic, Santa Barbara, Calif.

1. Janney, N. W.: J. A. M. A. **70**:1131 (April) 1918.
2. Folin, O., and Wu, H.: J. Biol. Chem. **38**: No. 1 (May) 1919.
3. Benedict, F. G.: Boston M. & S. J. **178**:667 (May) 1918.
4. Barrett, A. M.: Arch. Neurol. & Psychiat. **2**:628, 1919.

laboratory without breakfast and rest at least thirty minutes before the test is begun. Three readings of ten minutes each were taken in all cases, and the average of the results used as a basis of calculation. Respiratory quotient was assumed to be 0.82.

A brief summary of the cases follows: Data not mentioned may be regarded as normal. Summaries of the differential blood counts and glucose tolerance tests are given in Tables 1 and 2.

CLINICAL DATA

Included in the present study were members of three thyroid families presenting cases of unusual interest. Thyroid family 1 proved to have four members suffering from thyroid disease. This family came under observation because three young children had failed to grow normally. The remote family history was negative for goiter, obesity, growth anomalies, but always highly neurotic. The children were reared in the best social environment.

CASE 1 (No. 1938).—Female, aged 6 years (sister of Cases 2 and 3 [Nos. 1939 and 1940]); a child of highly neurotic disposition; masturbation at 1 year; growth which was normal until fourth year, but only three pounds had been gained in the last two years. Few physical abnormalities. Facial expression normal. Has eczema and very rough, dry skin. Perspiration deficient. Difficultly palpable thyroid. Slightly protruding abdomen. Thin, under-developed nails and tendency to decreased muscular tonus. Metabolic studies show inability to gain weight on large amounts of assimilable food and loss of nitrogen in urine and feces. The preceding, together with the fact that two other members of the family are quite definitely thyroid cases, and resumption of normal growth, general health and spirits after administration of ¼ grain thyroid extract, seems sufficient proof that the growth anomaly is due to a mild degree of hypothyroidism (Fig. 3).

Diagnosis.—Hypothyroidism; nervous type.

CASE 2 (No. 1939).—Male, aged 5 years (brother of Cases 1 and 3 [Nos. 1938 and 1940]); birth weight, 9 pounds and 11 ounces; walked at second year, but not well. Present weight, 43 pounds. Talked at twenty-second month. Colitis with bronchitis and asthma in second year. Tonsils and adenoids removed because of hypertrophy. Eczema at 1 year. Otitis media at 3 years. Present height 43 inches. Cannot reason properly; has defective memory and unreasonable temper. Has gained 3 pounds during past two years. Appetite poor.

Physical Examination.—Facial expression not suggestive of cretinism. Patient somewhat thin, hair normal, eyebrows very scanty. Nose and eyes small; nares flaring. Skin markedly rough and dry. Thyroid not palpable. Abdomen definitely protuberant. Stands with feet wide apart. Gait slightly waddling, gets up awkwardly and weakly from floor. Knee jerks normal, no paralysis or atrophy. Pulse 70. Roentgenologic studies showed marked underdevelopment of base of skull, facial bones and carpi. Metabolical studies show inability to gain weight on large amounts of assimilable food and loss of nitrogen in urine and feces (Table 4).

Diagnosis.—Hypothyroidism; nervous type. Improvement on thyroid treatment.

CASE 3 (No. 1940.—Female, aged 2 years (sister of Cases 1 and 2 [Nos. 1938 and 1939], daughter of Case 4 [No. 1936]). Sleeps very poorly. Screams and becomes nervous at the slightest provocation. Birth weight, 9 pounds. Early growth normal. Sept. 8, 1918, weighed 24 pounds 2½ ounces; Jan. 29, 1919, weighed 29 pounds 2½ ounces; Sept. 16, 1919 weighed 30 pounds 6 ounces. Child presents appearance of malnutrition, but no symptoms of cretinism. Physical examination otherwise negative. Certain diagnosis impossible, owing to tender age preventing laboratory tests and control of thyroid treatment. Child probably hypothyroid, in view of family history.

CASE 4 (No. 1936).—Male, aged 55; no complaint. Consulted because of thyroid condition of children (Cases 1, 2 and 3 [Nos. 1938, 1939 and 1940]). No goiter. Has tendency to gain weight of late. Average weight 128 pounds; present weight 135 pounds. Has had noticeable tremor of hands for past two years, following strain of war work. Nervous by nature.

Physical Examination.—Small stature. Manner highly neurotic; body movements very active. Hair of head dry and scanty. Exophthalmos quite marked, but special eye signs negative. Eyebrows, eyelashes, axillary and pubic hair very scanty. Skin normal. Definite fine tremor of extended hands. Panniculus slightly increased. Examination otherwise negative, including thyroid gland. Basal metabolism plus 11 per cent.

Diagnosis.—Thyroid dystrophy.

Thyroid Family 2 has four members, in two of whom hypothyroidism had been diagnosed previously. None of these patients shows typical symptoms, one member, an older brother (aged 17), of Case 6 [No. 1980], presents only a saddle nose and swelling of under lids; otherwise no mental or physical signs of hypothyroidism. His basal metabolism is normal, but he nevertheless tolerates a greater amount of thyroid than does a normal individual. The aunt of this patient shows myxedematous symptoms. Case 6 [No. 1980] proved most interesting, a total deficiency of the outer two-thirds of the eyebrows leading to examination and diagnosis. After three weeks of thyroid treatment, elliptical hyperemic areas developed about the location of the eyebrows which a little later grew to normal proportions. The cases of the mother and this son are as follows:

CASE 5. (No. 1979).—Female, aged 48; divorced; complaining of headaches, feeling of "tenseness," mental dullness, bad memory, loss of power of concentration. Was very stout child. Weighed 130 pounds at 20. Present weight 140 pounds. Menstrual history normal. Three children, one definitely hypothyroid (Case 6 [No. 1980]); another showing hypothyroid symptoms. Always very subject to infections.

Physical Examination.—Flushes easily; tâche cerebralé present. Expression decidedly apathetic. Speech slow. Mentality noticeably sluggish; ready forgetfulness during questioning. At times irritable without provocation. Panniculus largely increased, hands and feet cold, rather short and thick, no padding. Nails frail, skin dry and harsh. Hair of head scanty and dry. Axillary and pubic hair scanty. Eyebrows defective at outer third. Lids drooping, eye signs negative. Legs nearly hairless. Reflexes normal. Thyroid palpable, normal size and texture. Initial basal metabolism, —19 per cent.

Diagnosis.—Hypothyroidism. Improved on thyroid therapy.

Case 6 (No. 1980).—Male, aged 12; referred by hypothyroid mother (Case 5 [No. 1979]). No complaint. Healthy looking boy. A little dull at school. Skin dry. Pulse 78. Genitals undeveloped. Hands and feet cold, short and stubby; slightly cyanotic. Eyebrows very scanty, outer two thirds absent. Saddle nose. Slight fulness of lower lids. Thyroid scarcely palpable. Panniculus normal. Patient has had uvula amputated because of unusual size. Initial basal metabolism, —26 per cent.

Diagnosis.—Hypothyroidism. Improvement on thyroid therapy.

Thyroid Family 3 presented a number of abnormal individuals, only two of whom could be obtained for clinical study (Cases 7 and 8 [Nos. 1906 and 1907]). All members of this family have a tendency to obesity. One sister, not reported, weighed 225 pounds at 50, then developed exophthalmic goiter; another suffered from dementia praecox and one nephew had periodic insanity. The interesting feature in this family is the presence of degenerated thyroids associated with insanity among its members. This association will be alluded to later in the article.

Case 7 (No. 1906).—Female, aged 66 (sister Case 8 [No. 1907]); school superintendent. Complaining of obesity, swelling of ankles, slight dyspnea, constipation, exhaustion. Dyspnea very slight, with some palpitation. Menopause at 38, sudden termination.

Physical Examination.—Pulse 56; facies heavy, but is mentally quite alert; obese; hair gray, dry; eyebrows, axillary and pubic hair scanty; skin dry, cool, showing considerable atrophy. Extremities cold. Arcus senilis marked; eye movements normal. Thyroid not palpable. Postcervical fat pad. Systolic murmur at apex; abdomen rotund, large amount of subcutaneous fat. Slight edema of ankles, large fat pads around both ankles, especially on inner surfaces. Basal metabolism, —23 per cent.

Diagnosis.—Hypothyroidism; mitral insufficiency. Definite improvement on thyroid therapy.

Case 8 (No. 1907).—Female, aged 70 (sister of Case 7 [No. 1906]); widow. History of high blood pressure, complaining of dizziness and obesity. Has gained weight since 45 years of age, from 130 pounds to 196 pounds. Present weight, 174 pounds. Some swelling of ankles for four or five years. Severe headaches before 50. Always very subject to infections.

Physical Examination.—Short, obese. Skin shows senile or endocrinic atrophy, very dry; eyebrows scanty, especially outer third. Thyroid scarcely palpable. Dorsal cervical padding. Heart enlarged. Abdomen relaxed, panniculus greatly increased. Extremities cold, short and stubby. No hair on legs. Definite padding on inner surfaces of both ankles. No edema. Blood pressure, 190/80. Blowing systolic murmur over aortic area; accentuated second pulmonic and aortic sounds. Urine showed faint cloud of albumin; no casts; low specific gravity. Basal metabolism and sugar tolerance could not be studied.

Diagnosis.—Hypothyroidism; chronic diffuse nephritis. Improved under thyroid therapy.

Case 9 (No. 19104).—Male, aged 56; complains of nervousness, obesity, nervous exhaustion. Was very fat child; weighed 218 pounds at 14; 265 pounds at 17. Mentality keen. Always nervous. "Nervous breakdown" at college with severe melancholia. Second breakdown at 35 years. Became impotent at 35. Declared "thyroid case" at 48, and became potent after three weeks thyroid treatment. Present weight, 205 pounds.

Physical Examination.—Large frame, small bone development. Hair of head dry and scanty. Earlobes fused to head. Brown cutaneous pigmented areas. Subcutaneous tissue firm in areas, soft in others. Skin of back shows patches of roughened epidermis. Eyebrows scanty in outer third; eyelashes very scanty. Slight von Graefe's sign, pupils sluggish to light. Thyroid definitely palpable (small for size of patient). Abdomen obese. Feet show padding on plantar surfaces. Reflexes normal. Initial base metabolism, —20 per cent.

Diagnosis.—Hyperthyroidism. Recovery under thyroid therapy.

CASE 10 (No. 1998).—Female, aged 28; nurse. Headaches, obesity. Very fat child, weighed 120 pounds at 12 years. Present weight, 175 pounds. Height, 4 feet 11 inches. Menstruation irregular, very painful. Always nervous and emotionally unstable; recurring headaches, producing nausea and vomiting.

Physical Examination.—Short, heavy, obese. Stubby hands and feet. Definite fat pads at back of neck, over deltoid and inside of knees. Skin over upper extremities thick, harsh and cold. Hair normal. Thyroid not palpable. Initial basal metabolism, —14 per cent.

Diagnosis.—Hypothyroidism. Headaches immediately relieved by thyroid treatment.

CASE 11 (No. 1960).—Female, aged 40; nurse; exophthalmic goiter at 17. Thyroidectomy with removal of right lobe and isthmus at 31, at which time she was extremely nervous, and had some tremor and exophthalmos. Weight, 130 pounds. Subsequent uncontrollable gain in weight to 160 pounds, with swellings about eyes, mental dulness and lassitude.

Physical Examination.—Pulse slow; panniculus greatly increased; skin cold, harsh in spots; hair dry and coarse; hands and feet thickened; eyes slightly prominent, otherwise normal; nails brittle; typical myxedematous expression; puffiness of lower lids. No thyroid tissue palpable. Reflexes faint but present. Initial basal metabolism, —38 per cent.

Diagnosis.—Cachexia strumipriva. Cured by thyroid therapy.

CASE 12 (No. 1954).—Female aged 45, no vocation; general ill health, dizziness, poor memory and depression. Surgical menopause after oophorectomy eight years ago. Dizziness noticeably increased recently. Swimming sensations when lying down. No really definite complaint.

Physical Examination.—Panniculus greatly increased; enormously over abdomen. Hands cold, nails frail; hair dry and brittle; slight trembling of lids on closing. Flushes easily. Thyroid normal size; slight cutaneous atrophy; reflexes normal; pulse, 72. Initial basal metabolism, —16 per cent.

CASE 13 (No. 1991).—Female, aged 40; complaining of headaches and malaise. General health poor for five or six years. Headaches frontal and left side, sometimes generalized. Subject to infections. Five years ago patient became melancholic, irritable and nervous. Began taking thyroid extract on own initiative. Condition improved. Has taken 1 or 2 grains daily for four or five years. Gained 10 pounds in ten days one year ago while without thyroid. Marked improvement under controlled treatment.

Physical Examination.—Pulse, 60; height, 5 feet 3 inches. Panniculus greatly increased. Hands and feet small and stubby. Bone skeleton heavy for stature. Some cutaneous atrophy at knees and elbows. Skin dry, harsh and scaly. Hair dry. Eyebrows normal; eye signs negative. Tongue soft and small. Definite padding at base of toes and pedal surfaces. Reflexes normal. Initial basal metabolism, —11 per cent.

Diagnosis.—Hypothyroidism. Cured by thyroid therapy.

CASE 14 (No. 19112).—Female, aged 58; no occupation; dragging sensation in abdomen; fits of depression, melancholia. Tendency to gain in weight. Easily upset by trivial household worries. Complete hysterectomy thirty years ago. Mother and brothers had no eyebrows, and hair of body was very scanty.

Physical Examination.—Facial expression mildly myxedematous. Mentally active, but patient very irritable and complains of inability to concentrate thoughts. Panniculus slightly increased. Eyelids tremble slightly on closing. Eyebrows very faintly developed. Earlobes fused to head; slight puffiness of lower lids. Thyroid palpable. Slight padding of supraclavicular fossae. Abdomen relaxed, panniculus greatly increased. Extremities cool, skin very smooth, dry; nails frail and toe-nails quite atrophic. Definite cutaneous atrophy at elbows and lower limbs. Initial basal metabolism, —19 per cent.

Diagnosis.—Hypothyoridism. Improvement on thyroid treatment (see discussion).

Case 15 (No. 19101).—Female, aged 27; complaining of physical exhaustion. Grew normally as child. Growth terminated at 16. Much smaller in stature than other members of her family. Mentality exceptionally keen. Perspires excessively, especially arms. Of lively disposition and quick in movements. Best weight, 112 pounds; present weight, 106 pounds. Height, 5 feet.

Physical Examination.—Pulse, 80, but tachycardia on slight provocation. Patient flushes easily. Hair falling recently. Slight exophthalmos for past three years, with Stellwag sign, but no other special signs. Thyroid slightly increased in size. Palms moist. Tongue large; heavy mouth and lips. Skin normal texture, except for coarse areas over shoulders. Cutaneous pigmentation at elbows, axillary and inguinal folds. Uterus abnormally small. No other signs of hypothyroidism than noted. Basal metabolism, plus 5 per cent.

Diagnosis.—Thyroid dystrophy, tending to thyrotoxicosis.

Case 16 (No. 1964).—Female, aged 37; no occupation; complaining of periodical enlargement of the thyroid and physical exhaustion. Glandular enlargement occurs every three or five days, lasts from three to four days, and disappears. No relation to exercise, mental condition, or menstruation. Patient not markedly nervous nor annoyed by trifles. Psychoneurotic and emotionally unstable. Patient has gained 15 pounds in last year.

Physical Examination.—Does not look her age. Neurotic expression. Eyebrows normal. Eyelids tremble on closing. Corneal reflex dull. No other eye signs. Nares dilated, lips full. Mouth large in proportion to face. Tâche cerebralé present. Flushes easily. Tongue voluminous and fissured. Thyroid, little, if any, increase in size. Abdomen obese. Patient's movements suggestive of myxedema. Extremities cool, skin extremely coarse in places, no hair on hands; skin of thighs coarse; hands short and broadened. Distinct tremor of thyroid type. Thickened subcutaneous tissue. Reflexes normal. Basal metabolism, —14 per cent.

Diagnosis.—Thyroid dystrophy, with hypothyroid tendency. No treatment.

Case 17 (No. 20213).—Female, aged 51; no occupation; complaining of enlargement of neck. Enlargement began six years ago, accompanied by increased pulse rate and slight prominence of eyes. No noticeable tremor or nervousness. No mental disturbance. Subject to tonsillitis in youth. Articular rheumatism and hay-fever in early life. Always over weight, from childhood on. Highest weight, 180 pounds; present weight, 170 pounds. Subject to headaches.

Physical Examination.—Large frame, rather heavy features; hair good condition, eyebrows scanty; flushes and perspires easily (recent development). Suggestion of exophthalmos and Stelwag's sign. Puffiness of lower lids. Temporal veins congested. Breathing stridulent, occasional brassy cough. Slight saddle nose. Tongue large, shows definite tremor. Both lobes of thyroid markedly enlarged; right lobe larger and more dense than left. No fixation. Heart and lungs negative. Abdominal panniculus greatly increased. Hands short and stubby, moist palms, fine tremor. Nails small, frail and brittle. Legs smooth and hairless. Knee jerks slightly exaggerated; other reflexes normal. Hair distribution normal, but axillary and pubic hair very scanty. Basal metabolism, —10 per cent.

Diagnosis.—Thyroid dystrophy. Treatment expectant. Lobectomy advised, if pressure symptoms grow worse.

DIAGNOSIS OF HYPOTHYROIDISM

Relative Importance of Clinical Symptoms.—In seventeen consecutive cases we have encountered but one clearly cut case of cachexia strumipriva presenting the entire classical syndrome of myxedema. All the other cases, with exception of four cases of dysthyroidism, were of the masked type of hypothyroidism originally described by Hertoghe. The strong familial tendency of thyroid disease is illustrated by the finding of three thyroid families among our series.

In order to determine as far as possible with our limited material what points in the ordinary clinical examination are most important in the diagnosis of thyroid deficiency, Table 1 has been prepared, in which the relative frequency of the symptoms is noted. The following may be mentioned as important from the standpoint of occurrence being found in more than 50 per cent. of cases: history of obesity, particularly in early life; mental symptoms, marked liability to contract infections, hair anomalies, dry, harsh skin with pigmentation and atrophy; cold extremities and cold skin generally, decreased size of thyroid, subnormal temperature, pulse and respiration.

TABLE 1.—Relative Frequency of Thyroid Symptoms

	Cases or Per Cent.
Ancestry with history of glandular disturbances or obesity	3 or 17
Personal history of tendency to obesity	12 or 70
Mental symptoms	11 or 64
Including:	
1. Mental dulness	3 or 17
2. Irritability	7 or 41
3. Headache	5 or 29
4. Melancholia	4 or 23
5. Poor memory	3 or 17
Anomalies of development	5 or 29
Liability to infection	9 or 51
Exhaustion or lassitude	6 or 35
Physical signs:	
Hair anomalies	12 or 70
Dry, harsh skin, with pigmentation or atrophy	13 or 76
Frail, poorly developed nails	6 or 35
Cold extremities and skin	9 or 52
Stubbiness of hands and feet	7 or 41
Dull expression, heavy lips, large tongue, saddle-nose	7 or 41
Increased panniculus	12 or 70
Thyroid:	
Enlarged	2 or 11
Normal	4 or 25
Decreased	11 or 68
Fat pads:	
Dorsum of hands, feet, supraclavicular fossa and back	7 or 41
Subnormal temperature, pulse and respiration	14 or 81

With regard to the relative importance of special symptoms, slowing of pulse, respiration and a subnormal temperature seem to have particular significance. These changes were present in 81 per cent. of this series, including very mild cases. They seem at times to represent possibly the only signs of latent thyroid deficiency. For example, Case 199,

not included in the detailed clinical study, may be cited. A lady complained for years of nervous and physical exhaustion on slight provocation. Careful medical review of her case recorded a negative family history, with exception of two sisters having a similar condition and slow pulses. Physical examination, essentially negative, with exception of a slow pulse, 54 minute. The basal metabolism, —11 per cent., was at the lower level of normal, but suggested possible slight hypothyroidism. A trial of 0.4 mg. thyroxin per day, resulted in a few weeks in general improvement. The pulse and the basal metabolism rose to normal. This therapeutic test would seem to justify a diagnosis of latent hypothyroidism in this case.

Examination of the thyroid may be quite misleading. Twenty-five per cent. of our cases exhibited thyroid glands normal to physical examination. It must likewise be remembered that involution of the thyroid takes place as age advances. A small thyroid in an elderly individual is per se no evidence of myxedema, unless the gland be scarcely palpable, which should arouse suspicion. Family history suggestive of thyroid disease, fat padding, marked frailty and brittleness of the nails, growth anomalies, though less frequent, are, when present, of diagnostic importance. Sufficient attention is not always given to examination of the hair and nails, which are very often anomalous in thyroid disease. Dystrophies of hair and nails are nearly always present in myxedema, thus bespeaking their relative importance. In our series their less frequent occurrence may be ascribed in many instances to the mildness of the thyroid deficiency.

Mental symptoms, as a result of latent hypothyroidism, are more common than usually accepted by the neurologist or internist. Very few hypothyroid cases show, if carefully observed, a normal mentality. We look too often for mental lassitude, forgetting that symptoms of mental irritability are also very frequently met with. Sometimes the picture is only that of neurasthenia. It may, however, go on to insanity. In one of our thyroid families, one individual had dementia praecox, another periodic insanity. Such cases may not be on a thyroid basis, but only accompanying degenerative phenomena. Simple failure of memory and the power of concentration are frequently met. A very interesting thyroid family of sixty-two members has recently been reported by Barrett,[4] marked anomalies of the hair and nails being present in many members, together with mental conditions and other signs of degeneration.

The Differential Blood Picture.—The blood in thyroid disease, as first pointed out by Mennacher, tends to revert to the fetal type which is characterized by a preponderance of the lymphocytic elements, with corresponding decrease of the polymorphonuclear cells, together with a mononucleosis. This has been clearly demonstrated by the work of

Kocher and his pupils, Falta,[5] McCarrison,[6] and others. These changes come on early and are very persistent, occurring in both hyperthyroid and hypothyroid cases. It seems strange that previously the diagnostic significance of the differential count in masked hypothyroidism has not been sufficiently emphasized. In all but one case of one series, this blood picture was present (Table 2) the average of thirteen adult patients being: polymorphonuclears, 55.1 per cent.; lymphocytes, 36.1 per cent.; large mononuclears, 3.53 per cent. The mononucleosis is rather constantly present. In children more than the normal lymphocytosis, also mononucleosis, is found. We have not observed eosinophilia, except in one hypothyroid case after thyroid treatment. Eosinophilia has been reported in hyperthyroid cases.

TABLE 2.—Differential Blood Counts in Thyroid Cases*

Case No.	Polymorphonuclears	Lymphocytes	Large Mononuclears	Eosinophils	Other Elements	Remarks
1900	62	33	4	0.3	0.7	Before adequate treatment
	56	38	3	1	3	After treatment
1954	48	45	6.3	0.6	0.1	Before treatment
1906	55	38	4	1	2	Before treatment
19112	61	35	3	1	0	Before treatment
1996	49	49	1	0.3	0.66	Indifferent treatment
1980	65.3	32.3	1.6	0.6	0	Before treatment
1979	49	43	5	0.66	2.33	Some previous treatment
1907	63	32	4	1	0	Before treatment
1958†	42	55	1	1	1	Before treatment
1989†	49	49	1	1	0	Before treatment
19104	55	40	2	1	2	After long unsystematic treatment
1940†	30	67	1	2	0	Before treatment
1900‡	58	35	4.5	2.5	0.0	No treatment; thyroid dystrophy
19101	63	28	6.6	2	0.4	No treatment; thyroid dystrophy
19213	73	21	1	2	3	No treatment; thyroid dystrophy
Average of adult cases	51.1	36.1	3.53	0.99	1.09	

* Three hundred cells counted.
† Not included in average (children).
‡ Two hundred cells counted.

This same type of differential blood count is, unfortunately, not pathognomonic of thyroid conditions, as it is exhibited by various endocrine disturbances, such as Addison's disease and hypopituitarism. It seems possible, however, from the finding of an eosinophilia in a number of hypopituitary cases by Falta and co-workers, that this might prove a differential point in diagnosis from hypothyroidism on further observations in a large series of cases. In cases of uncertain symptomatology, we have found the differential blood count to be very helpful as corroborative evidence, together with the basal metabolism and the blood sugar tolerance curve, in determining the diagnosis of thyroid disease.

5. Falta, W.: Diseases of Ductless Glands, Ed. 2, Philadelphia, 1916.
6. McCarrison, R.: The Thyroid Gland in Health and Disease, New York, William Wood & Co.

THE BASAL METABOLISM

No more interesting example of the successful application of a purely experimental method to modern clinical medicine exists than in the case of basal metabolism. But a few years ago determinations of the basal metabolism were necessarily carried out only in a very few physiologic laboratories, sufficiently well supported to bear the enormous expense of the upkeep of a respiration chamber. The reduction of this method to clinical utility by the recent introduction of smaller apparatus, especially the Benedict portable form, is now being followed by its coming into general use. We are indebted to Plummer [7] for the clinical application of basal metabolic study on a large scale. This method has the immense advantage of being entirely objective, and is thus peculiarly applicable to borderline cases.

In common with Magnus-Levy, DuBois,[8] Means and Aub,[9] we have also found this method the best laboratory aid to diagnosis of thyroid conditions we now possess. It has been applied in every case of the present series. Enthusiastic as we are in its use, attention should be called, however, to certain limitations for avoidance of errors.

The estimation of basal metabolism is a method of precision. Even with the simplest possible technic, the method must be constantly used to insure accuracy. There will be shown on the side of clinicians, perhaps, a too great dependence on this single laboratory procedure, especially when in some cases the technic may be fairly questioned. In using the small Benedict apparatus, only variations in basal metabolic determinations of more than 10 per cent. from the normal can be considered of significance. A moderate increase in basal metabolism above 10 per cent. may be due to other factors, such as fever, faulty determinations due to movement of patient, etc., as well as to hyperthyroidism.

Variations in the metabolic rate are not necessarily due to thyroid disease. Thus we have observed a rate of —15 per cent. in hypopituitarism. Depression of the metabolic rate from 20 to 40 per cent., is usually clearly indicative of thyroid deficiency. However, it must again here be remembered that patients in lowered states of nutrition may show greatly depressed metabolic rates. Thus, among diabetics without thyroid disease, we have observed rates as low as —35 per cent. The metabolic rate may also fail to serve as an exact criterion in early cases of hyperthyroidism, and even exophthalmic goiter,

7. Plummer, H.: Proceedings of the American Medical Association, New Orleans, 1920.
8. DuBois, E. F.: Arch. Int. Med. **17**:915 (June) 1916.
9. Means, J. H., and Aub, J. C.: Arch. Int. Med. **24**:404 (Oct.) 1919.

although in the vast majority of cases, it is a great aid to the appreciation of the severity of the case as well as to therapy. Thus can be quoted a case of exophthalmic goiter recently observed by us showing thyroid hypertrophy, tremor of fingers and tongue, sympathicotonia, moist palms and soles, nervousness and excitability, exophthalmos, Stellwag's sign, the glistening eyeball, rapid pulse on slight provocation, loss of weight and strength, indeed, all the symptoms necessary for a diagnosis, yet the basal metabolism was 5 per cent. below normal. Sound clinical judgment should never be unduly influenced by laboratory methods.

THE BLOOD GLUCOSE TOLERANCE TEST

This test, as applied to the thyroid, is founded on substantial experimental basis [10] which may be cited in abstract. Normal values for the blood sugar and the blood glucose curve were obtained by us in a series of dogs. The thyroid glands were then removed, the presence of intact parathyroids being determined by subsequent postmortem. After complete recovery, the blood sugar level was again observed and blood sugar tolerance curves established. In every case marked hypoglycemia and a remarkably flattened curve resulted with a delayed return of the blood glucose to the normal.[11]

It was therefore expected that a hypoglycemic delayed curve would be found typical for hypothyroidism in the human being, especially as subsequent studies demonstrated marked hypoglycemia in Addison's disease and Frölich's syndrome, as well as in muscular dystrophy, which is probably a hypofunctional endocrinic syndrome.[12] Our present studies, however, make it very doubtful whether a hypoglycemic hypothyroid blood glucose curve, as distinguished from a possible hyperglycemic hyperthyroid curve, can be really established. As is shown in Figure 1, hypoglycemic, hyperglycemic and normal blood sugar curves have been found in our hypothyroid cases. Though hyperglycemic curves may be due in part to thyroid treatment, these differences can scarcely be ascribed to technic, as the quantitative precautions now in general use were adopted.[1] From our experience, we may merely conclude that hypoglycemia and a lowered blood sugar curve are more common in hypothyroidism than in hyperthyroidism. Here we would call attention to a point usually insufficiently emphasized in blood sugar studies. As important at least as the height of the hyperglycemic response to sugar ingestion is the time required after ingestion of glucose for return to the normal blood sugar level. Individuals

10. Janney, N. W.; Goodhart, S. P., and Isaacson, V. I.: Arch. Int. Med. **21**:188 (Feb.) 1918.
11. Janney, N. W.: Arch. Int. Med. **22**:162 (Aug.) 1918.
12. Janney, N. W., and Isaacson, V. I.: Arch. Int. Med. **22**:160 (Aug.) 1918.

with sound endocrinic organs show remarkable constancy in this regard, if a renal factor be excluded. A return to the normal level may be expected in from one and one-half to two hours from the sugar meal. Thyroid cases in most instances show distinctly delayed curves.

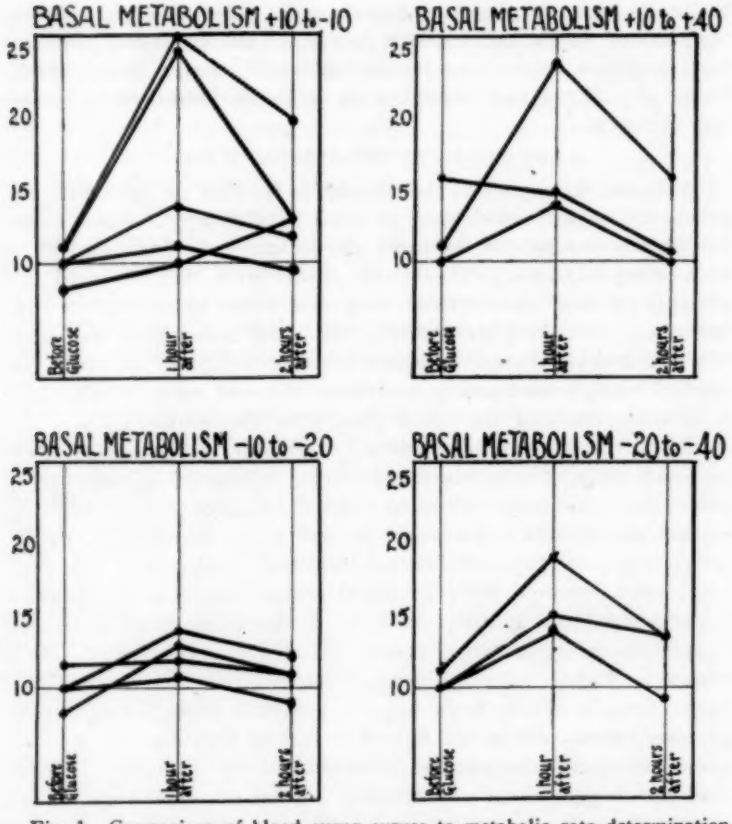

Fig. 1.—Comparison of blood sugar curves to metabolic rate determination in thyroid cases. Cases represented in four groups according to basal metabolic rate. Vertical numbers indicate blood sugar in parts per 10,000. The vertical lines refer to hours.

This, then, seems the one thing characteristic. The experimental recorded evidence quoted above would tend to give added importance to this observation, which has also been reported by Denis,[13] Lueders [14]

13. Denis, W., Aub, J. C., and Minot, A. S.: Arch. Int. Med. **20**: 964 (Dec.) 1917.

14. Leuders, C. H.: Arch. Int. Med. **24**:432 (Oct.) 1919.

and others in describing blood sugar studies. We are, however, confronted with the fact that other endocrinic dystrophies give likewise abnormal glycemic tolerance curves, showing this and all other abnormalities observed in thyroid diseases. One is, therefore, forced to the conclusion that an abnormal blood sugar tolerance curve (whether hyperglycemic, hypoglycemic, or merely a delayed return to normal) is to be regarded solely as a general indication of endocrine disturbance. Inasmuch as the highest curves with glycosuria are observed in exophthalmic goiter, and the lowest curves accompanied by increased sugar tolerance in hypothyroidism, such modifications of the curves are more likely to occur in cases exhibiting respectively hyperfunctional and hypofunctional conditions of the thyroid. Of this, however, there is no certainty in any given case.

In Figure 1 a number of blood sugar curves from various thyroid cases are grouped according to the basal metabolic rate. It will be seen, as also recorded by McCaskey,[15] that there is no definite relationship between either the height or delayed response in these curves, and the metabolic rate. A case of thyroid dystrophy with normal metabolic rate gave a markedly high and delayed curve with glycosuria. A myxedema patient with a —40 per cent. metabolic rate gave a hyperglycemic curve, 10, 16, 13 parts per 10,000 at hourly intervals. In spite of all the foregoing criticism, the blood sugar curve is, however, not without its practical utility in endocrine cases. Instances have already been observed in which the gaseous exchange is normal but evidence of disturbance of metabolism was afforded by the abnormal blood sugar curve. This we have observed in dysthyroidism and in incipient cases of hyperthyroidism. A case of mild exophthalmic goiter exhibited a normal metabolic rate, but the blood sugar curve (8, 17, 13 parts per 10,000 at hourly intervals) showed both increase in height and markedly delayed return to the normal level. An accurate blood sugar curve can often be obtained when elaborate basal metabolic apparatus is not accessible. For these reasons we suggest that blood sugar curves be made in all doubtful cases of endocrinic disturbance in addition to metabolic determinations.

THE NITROGEN METABOLISM

In connection with the present research we have investigated, mainly for the great theoretical interest connected with the same, the nitrogen metabolism in two cases of mild hypothyroidism in children of thyroid Family 1 (see above). The diets employed were of normal mixed constituents and in slight excess of the requirements for

15. McCaskey, G. W.: J. A. M. A. **73**:243 (July 26) 1919. New York M. J. **110**:607 (Oct.) 1919.

each child at the age when the experiments were undertaken. Both children were retarded in growth, and showed slight clinical signs of hypothyroidism. Proper care was exercised in execution of this work by adopting the same technic as hitherto used in numerous nitrogen balance experiments. The results were striking (Tables 3 and 4). The child presenting the less severe clinical symptoms failed to gain even a gram of nitrogen in a week's time; the other, exhibiting more thyroid deficiency, actually lost nitrogen. Interesting experimental evidence of defective thyroid function was thus afforded.

TABLE 3.—Nitrogen Balance in Hypothyroidism, Case 1 (No. 1938)

Days	Weight in Kilos	Total Calories	Calories per Kilo	Food Nitrogen Grams	Urine N_2	Feces		Nitrogen Balance
						Grams	Total N_2	
1	25.7	1350.3	67.8	10.1	8.9	.....	.....	
2	25.0	1709.8	67.8	9.9	12.6	.....	.....	
3	25.0	1759.5	70.3	10.1	9.16	.....	.....	
4	25.2	1960.3	77.6	11.0	10.84	.....	.....	
5	25.4	1966.2	77.8	10.2	9.2	.....	.....	
6	25.4	1635	64.3	11.8	7.56	.....	.....	
7	25.4	1750	68.9	11.1	10.84	.....	.....	
Total	....	.....	....	74.2	69.11	95.8	4.48	plus 0.66 gm.
Daily average	....	.....	....	10.8	9.88	....	0.63	plus 0.093 gm.

TABLE 4.—Nitrogen Balance in Hypothyroidism, Case 2 (No. 1939)

Days	Weight in Kilos	Total Calories	Calories per Kilo	Food Nitrogen Grams	Urine N_2	Feces		Nitrogen Balance
						Grams	Total N_2	
1	19.8	1451.2	75.9	8.04	9.7	.....	.....	
2	19.7	1503.8	75.9	7.8	11.6	.....	.....	
3	19.9	1523.5	77.3	9.6	12.58	.....	.....	
4	19.8	1957.3	98.1	9.9	6.25	.....	.....	
5	19.8	1959.3	91.3	10.1	6.25	.....	.....	
6	19.8	1550.7	78.0	10.1	10.5	.....	.....	
7	19.8	1526.5	77.0	8.1	6.48	.....	.....	
Total	....	.....	....	63.64	63.36	90.8	5.959	—5.95
Daily average	....	.....	....	9.09	9.05	....	0.835	—0.195

We have previously made use of the nitrogen balance to demonstrate the synthetic action of the thyroid gland on protoplasm, but did not recommend this as a clinical method for general use. In spite of this, in recent literature our nitrogen balance experiments have been criticized as cumbersome and not to be preferred to the basal metabolism, etc., all of which would, of course, be applicable, had we advanced nitrogen balance experiments as a clinical test in thyroid disease. In the examples just described, however, the nitrogen balance work served a direct clinical aim as the results obtained enabled

us to verify the obscure diagnosis of possible hypothyroidism in children too young for metabolic basal rate determination with the means at hand.

The Therapeutic Test in the Diagnosis of Hypothyroidism.—Although put in the background by the introduction of the laboratory aids to diagnosis, the therapeutic test is not to be forgotten in doubtful cases. Recent exact studies have, however, emphasized that there is a very definite thyroid requirement in each case of thyroid deficiency. If no definite therapeutic effect therefore, be observed, either too little or too much thyroid hormone may have been administered and the thyroid factor remain undiscovered. Full effect only takes place slowly. A trial of several weeks, at least, is usually necessary before judgment can be passed.

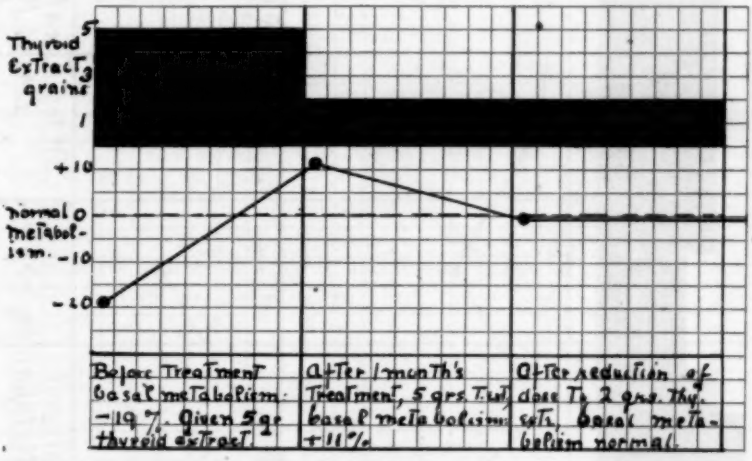

Fig. 2.—Treatment of hypothyroidism controlled by basal metabolism determinations, Case 5 (No. 1979).

TREATMENT OF HYPOTHYROIDISM

It has been our custom for the past year to control the treatment of cases of hypothyroidism by basal metabolism studies. With judicious thyroid treatment, it is surprising with what rapidity the basal metabolic rate rises, accompanied with prompt improvement of all clinical symptoms. In most cases there is excellent parallelism between the clinical course and the basal metabolic rate, as Figures 2 and 3 indicate. Frequently a change in the metabolic rate will give, even before the clinical symptoms, a clear indication for change in dosage. This is, of course, of great advantage in treatment, as the

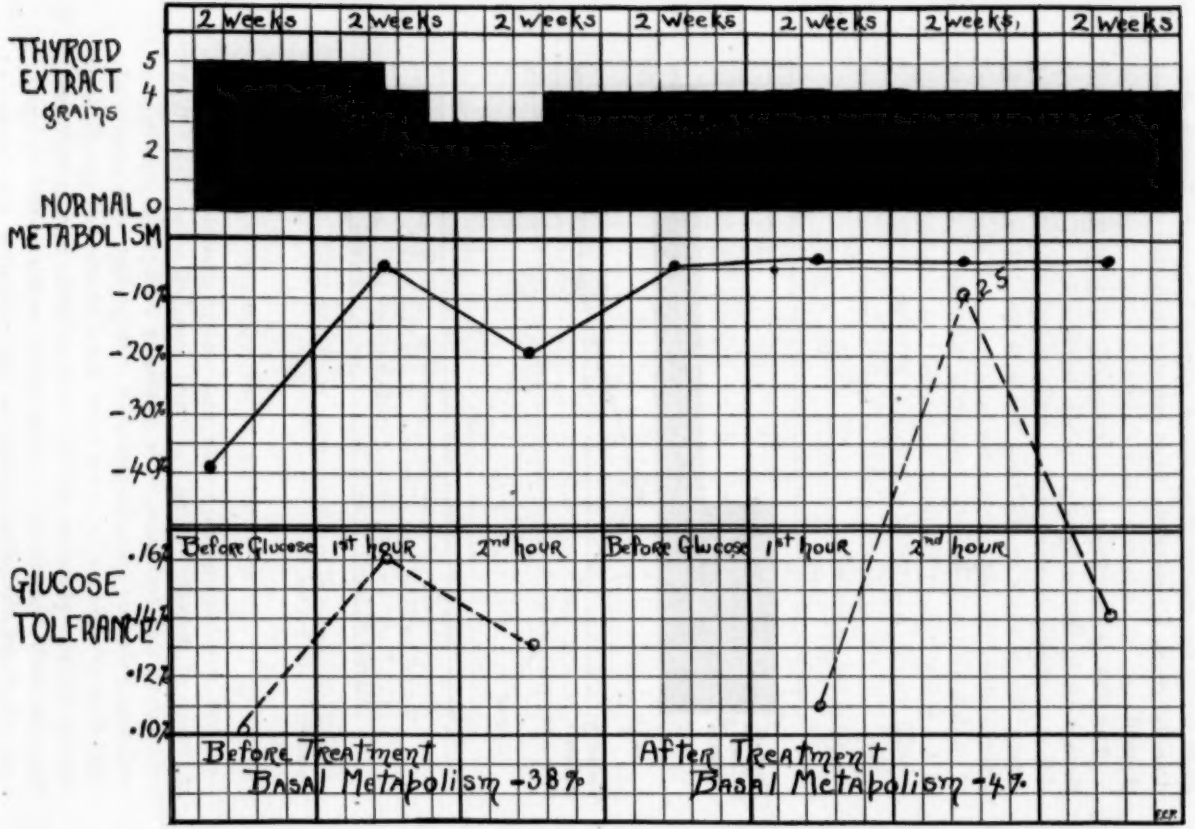

Fig. 3.—Treatment of cachexia strumipriva controlled by basal metabolism determinations, Case 11 (No. 1960).

personal factor of clinical judgment can be balanced by an exact physical method. Frequently, too, necessary variations in dosage can be controlled very accurately by metabolic rate determinations. We fear, however, that there will develop a tendency in the profession to put too great confidence in a normal metabolic rate. We have observed several cases of hypothyroidism in which thyroid caused a permanent return to the normal rate in short order, but clinical improvement, even after months of treatment, was not very satisfactory. The patients remained over weight, mentality was only partly improved, pulse and temperature subnormal. One must, of course, be mindful that the brilliant results of thyroid treatment in young children cannot always be duplicated in adult cases of many years standing, where skeletal and other organic changes must be regarded as permanent. Moreover, maximal improvement may require months or even years.

An interesting and discouraging group of cases in regard to therapy are the dysthyroid cases, previously discussed. In these borderline cases, as evidence accumulates, the basal metabolic rate will probably prove very helpful in indicating proper therapy. There is present a confusing complex of "hyper" and "hypo" symptoms, with basal metabolic rates which may be slightly above, below normal, or exactly at the normal level, due regard being given for the limits of error of the method. These cases showing decreased metabolic rates can properly be regarded as preponderantly hypothyroid and given thyroid treatment with expectancy of some improvement. For those showing normal or slightly increased rates, little can be done. A thyroidectomy would incur the risk of subsequent myxedema. Fortunately many of these cases are nonprogressive, some, indeed, being so little inconvenienced by their symptoms that their medical condition is only accidentally discovered.

Attention is called to two cases of hypothyroidism in women (Cases 12 and 14 [1954 and 19112]) having previously undergone complete removal of ovaries. In these cases the basal metabolism could not be brought to normal, although it improved on thyroid treatment, as symptoms of distress, palpitation and rapid pulse intervened even on small thyroid doses. The question of depression of the basal metabolism through loss of ovarian function is brought up by these findings.

Now that the thyroid hormone has been duly discovered, its chemical formula and characteristics, also physiologic effect, well established by the splendid researches of E. C. Kendall, it is advisable to use this preparation to the exclusion of all others depending more or less uncertainly on their thyroid hormone content. In 1917, Kendall presented us with an amount of thyroxin for metabolism study at the

Montefiore Hospital, New York. Further experience has been gained during the past year, when it has been in routine use. In all ways can we corroborate the findings of the Mayo Clinic as to the physiologic and clinical effect of thyroxin. Already put on a commercial basis, this preparation has the sole objection of being somewhat expensive at present.

GENERAL ASPECTS OF METABOLISM IN HYPOTHYROIDISM

There still remains considerable confusion with regard to modern conceptions of thyroid function. This, to a great extent, has been due to the antithesis between hypothyroid and the so-called hyperthyroid symptoms, as originally drawn by Kocher. On this has been built the hyperthyroid theory of exophthalmic goiter which has of late received additional impetus through the extension of basal metabolism determinations to clinical medicine. The increase in the metabolic rate has been accounted for by a purely dynamic theory of thyroid function. There are, however, many criticisms which render such a theory inadequate. These have been discussed by various writers, including one of the present in an earlier communication,[16] and are not pertinent to the present paper.

Study of the metabolism of hypothyroidism is, however, a greater aid to a clearer conception of thyroid function than the perplexing picture of thyrotoxicosis. Since Magnus-Levy's discovery of the decreased metabolic rate exhibited by cretins and myxedematous individuals, little experimental data had been added for a number of years. A reinvestigation of this subject was therefore undertaken in 1915 by one of us (N. W. J.) and his co-workers. This has been continued recently in our Santa Barbara Clinic, and additional data are included in the present report.

Since the decreased basal metabolic rate of cretins was accounted for by Magnus-Levy and others, as being caused by defective food absorption due primarily to alimentary tract sluggishness, exact feeding experiments [12] were undertaken which demonstrated that absorption of food from the intestines of athyroid animals was just as active as in normal animals. This led to further work on cretins, bringing out the fact that thyroid deficient patients failed to absorb as much nutritive material from the intestinal tract as normals, not because the alimentary function was necessarily deficient, but because such subthyroid individuals could not utilize digestive products in the normal manner, whether for fuel, tissue repair, or replacement purposes, *after* this food was assimilated by the intestinal tract. As was clearly demonstrated in prolonged metabolic experiments, when thyroid extract or thyroxin was administered in proper doses to these cretins, there

16. Arch. Int. Med. **22**:187 (Aug.) 1918.

ensued increased intestinal absorption accompanied by and probably required by increased ability to retain nutritive substances in the body and build them into new tissue. These data were obtained by following the nitrogen metabolism. With the demonstration that large amounts, even 100 per cent. more than previously, of nitrogen were retained on proper thyroid dosage, good, if indirect, evidence was secured that the thyroid synthetically controls the growth, probably also the constructive repair and replacement of tissue. The anabolic function of the thyroid gland on metabolism was thus experimentally demonstrated. Hitherto the thyroid was supposed to exert its effect on metabolism solely through the stimulation of catabolic processes as shown by increased nitrogen loss following the ingestion of thyroid

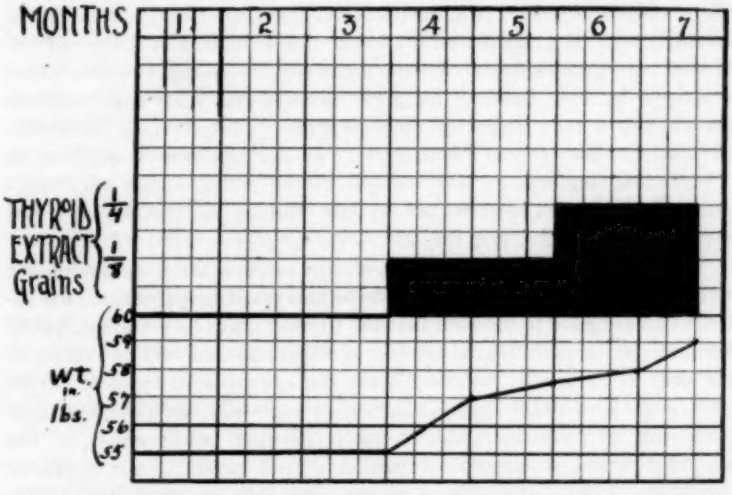

Fig. 4.—Resumption of growth in latent hypothyroidism of childhood with thyroid treatment, Case 1 (No. 1938).

material. Further experiments were therefore carried out demonstrating that the loss of nitrogen in this latter instance is a toxic loss, due to overgreat thyroid dosage.

The two nitrogen balance experiments reported in this paper are further illustrations of the inability of young hypothyroid individuals to properly utilize their food for fuel and growth, this inability being promptly recovered through thyroid treatment, as shown in the growth chart, Figure 4.

At first sight it seems difficult to correlate the well known catabolic action of the thyroid on metabolism with its synthetic function. Increased oxygen consumption and carbon dioxid output are generally

regarded as accompanying phenomena of combustion, i. e., catabolic processes. Therefore the thyroid has been looked upon as "the bellows which fans the fires of metabolism," and the basal metabolic rate as an exact guide to this function, as recently emphasized by Plummer's clinical studies. In the interpretation of basal metabolism studies, it must, however, be borne in mind that, in such, merely the total requirement for oxygen and the carbon-dioxid production of the body at a given time are so measured. *Oxygen, however, is required, as shown in recent intermediary metabolic studies, in anabolic as well as in combustive or catabolic processes.*[17] Though increase in the metabolic rate is at present the most exact laboratory aid in the determining of thyroid function, this method merely measures the degree of utilization or production of two essentially end products of metabolism. Mere reflection makes it indeed self-evident that, coincident with the establishment of an increased metabolic rate exhibited by a thyroid-fed cretin, true growth must of very necessity be taking place as characterized by the building of new tissue. Anabolic and catabolic processes are thus occurring coincidentally. Indeed, it is likely that the anabolic must be in the ascendant or the catabolic would lead to a persistent decrease in body weight of the cretin, which only takes place so far as is determined by the freeing of the organism of myxedematous tissue and fat.

Modern intermediary metabolic studies require, then, the acceptance of the coincident interaction of anabolic and catabolic processes. Prevalent evidence goes to indicate that the thyroid plays as yet a not clearly definite rôle in the intricate mosaic of these opposed sets of reactions, the end result of this function being the growth and replacement of tissue. It is probable that catabolism of ingested food products takes place for two reasons; one, to meet the fuel requirements of the organism; two, to supply intermediary split products for synthetic purposes. It is evident, from experiments showing the effects of ingested food on the thyroid gland of the fed animal, that the thyroid exert an influence in these preliminary catabolic reactions. Synthetic processes are, however, also carried out by aid of the hormones, of which the thyroid and pituitary are the most important, as is shown by clinical observations of the effect of these glands on growth and development. It would then undoubtedly render the whole conception of thyroid function vastly more lucid, were we to accept that the thyroid controls those metabolic processes necessary for the satisfaction of the heat, energy and regenerative requirements of the organism, including formation of new tissue.

17. Dakin, H. D.: Oxidations and Reductions in the Animal Body, New York, Longmans, Greene & Co.

The failure of parallelism between blood glucose determinations in thyroid patients and their basal metabolism (see above) is another indication that the metabolic disturbance in thyroid disease is more complicated than the metabolic rate alone would indicate. There is abundant evidence that the carbohydrate metabolism is abnormal in thyroid disease. There are also much data indicative that carbohydrate intermediary products are actually utilized in protein synthesis. The evidence supporting this view has been collected by Janney [18] and need not be quoted here.

Thus it may be mentioned, only for example, that respiratory quotient studies make it seem unlikely that the great disappearance of carbohydrate in active muscle is merely combustive. Furthermore, it is likely that side by side are two distinct processes taking place; one a degenerative, characterized by carbon dioxid output; another, regenerative, characterized by absorption of oxygen. In view of the anabolic action of the thyroid, as demonstrated by our metabolic experiments, and the close relationship of the thyroid to carbohydrate metabolism, it seems reasonable to draw the inference that the thyroid probably exerts, at least in part, its synthetic action on protoplasm by aid of the products of carbohydrate metabolism. This seems reasonable as it is now known that various amino-acids can be produced in vivo by the aminization of intermediate products derivable from glucose.

These views of thyroid function may now be applied to form a clearer conception of hypothyroidism. In this deficiency disease growth is inhibited; development is arrested; pathologic studies show extensive degenerative processes of the tissues; less food is absorbed from the alimentary tract, and what is absorbed, poorly utilized synthetically as well as combustively, as shown by the nitrogen metabolism studies; carbohydrate metabolism is abnormal; decreased excretion of the purin bodies and the appearance of urinary creatin takes place; metabolic activity is generally lessened, as shown by depressed basal metabolism. The entire picture of hypothyroidism is that of *impairment of tissue growth, regeneration and consequent degeneration* due to hormone hunger. It may, indeed, be possible that the great depression of basal metabolism observed in creatinism is partly, at least, of compensatory nature, energy being spared and all metabolic processes being inhibited on account of the inability to adequately utilize fuel and replacement materials. That such an explanation may apply seems likely from the results of basal metabolism determinations in inanition, in which the rates are quite as low as those of the most marked cases of hypothyroidism which have been recorded. All of these varying factors can be understood and correlated with the aid of acceptance of the view

18. Janney, N. W.: New York M. J. **107**:24 (May 4); 879 (May 11) 1918.

that the thyroid gland controls the metabolic processes, anabolic and catabolic, necessary for the required replacement and repair of the organism's cells, together with the production of heat and energy.

Thyroid therapy leads to stimulation of appetite, as more food can be assimilated for fuel and tissue construction purposes, increased absorption from alimentary tract, the increased combustion and synthetic utilization of the digestive products, as shown by the rise in basal metabolism and the increase in normal elimination of the purins, as their proper replacement and synthesis in the body is now possible; disappearance of creatin, as a sign of cessation of abnormal tissue breakdown. Regeneration thus takes the place of degeneration of tissues, growth and development return to normal.

SUMMARY AND CONCLUSIONS

Latent hypothyroidism is more frequent than generally supposed, as among eighteen consecutive thyroid cases, it was present in twelve, four cases being dysthyroidism and only one presenting classical myxedematous symptoms. Analysis of clinical data shows the following to be present in more than 50 per cent. of our series: history of obesity, particularly in early life, mental symptoms, marked liability to contract infections, hair anomalies, dry, harsh skin with pigmentation and atrophy, cold extremities and cold skin generally, obesity, decreased size of thyroid, subnormal temperature, pulse and respiration. Of all this data, lowered temperature, pulse and respiration occur most frequently, being found in 81 per cent. of our cases. Frequently but few symptoms may be present, diagnosis being impossible without laboratory methods. In the case of children with obscure symptoms, parents should be examined. Attention is called to the diagnostic value of lymphocytosis and mononucleosis in obscure thyroid cases.

The basal metabolic rate is of great value in diagnosis and treatment of hypothyroidism, but cannot be considered an absolute criterion.

The blood glucose tolerance test is abnormal in respect to the height of the curve and delayed return to normal level in the majority of cases of thyroid disease, but is only diagnostic of endocrine disease in general. There is no constant relation demonstrable between the blood sugar curve and the metabolic rate in thyroid disease. The blood glucose curve is, however, of confirmatory value in diagnosis of obscure thyroid cases.

Estimation of the nitrogen balance in two cases of obscure hypothyroidism showed an inability to retain nitrogen, this being further evidence of the synthetic function of the thyroid gland, as further developed in the theoretical consideration.

Treatment of hypothyroidism is best carried out with Kendall's thyroxin and controlled by estimation of the basal metabolic rate.

THE EFFECT OF ETHER ANESTHESIA ON THE ALKALI RESERVE *

AN EXPERIMENTAL STUDY

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The alkali reserve can best be determined by measuring the capacity of the blood for combining with carbon dioxid. Many of the observations to determine the influence of anesthesia on the alkali reserve have been made on patients after surgical operations. The alkali reserve may be decreased by the restricted diet or fasting preparatory to surgical operations, and the same condition is usually present in surgical shock, so that ether anesthesia produced experimentally in animals offers the advantage of eliminating these contributing factors.

Caldwell and Cleveland [1] studied the influence of different kinds of anesthesia on the alkali reserve in more than 100 patients, including fifty-five for whom ether anesthesia was used. In those latter cases they found a slight decrease, varying from 4.5 to 7.7 volumes per cent. in the combining capacity for carbon dioxid, but acidosis approaching dangerous proportions was noted only in the case of a diabetic patient who had acidosis before the operation.

Austin and Jonas [2] determined the carbon dioxid combining capacity of blood in sixteen patients after ether anesthesia for various surgical operations. The maximal decrease was 10 volumes per cent. and the lowest amount observed was 47 volumes per cent. The reduction seemed proportionate to the duration of the anesthesia and was maximal at the close of the anesthesia. When a decrease occurred, it apparently persisted with little alteration for about five hours.

Cannon [3] made a number of observations on soldiers who had been wounded in battle and were suffering from varying degrees of shock. In a series of nine cases of moderately severe shock, there was a decrease of approximately 12 volumes per cent. on the average, the maximal reduction being 16 volumes per cent. He found the decrease occurred rapidly when the capacity of the blood for combining with carbon dioxid was low (below 50 volumes per cent.) before the

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* From the Laboratory of Physiology of the University of Texas.

* Read before the Section on Pathology and Physiology at the Seventy-First Annual Session of the American Medical Association, New Orleans, April, 1920.

1. Surg., Gynec. & Obst. **25**:22, 1917.
2. Am. J. M. Sc. **153**:81, 1917.
3. J. A. M. A. **70**:531 (Feb. 23) 1918.

operation. The maximal decrease following operations when nitrous oxid and oxygen were used for anesthesia was 8 or 9 volumes per cent. Cannon expressed the opinion that the alkali reserve is below normal in shock, and that the reduction is in direct proportion to the degree of shock.

W. H. Morris [4] made a number of observations on patients before and after gynecologic operations. He also made a few experiments on dogs with prolonged ether anesthesia but without any surgical operation which could produce shock. He found that a reduction of the carbon dioxid combining capacity of the blood usually but not invariably occurs and that it does not bear any relation to the duration of the anesthesia. In one case in which the anesthesia only lasted twenty minutes, there was a decrease of 10 volumes per cent.; in another case lasting two hours for a hysterectomy, the decrease was 7.7 volumes per cent. The greatest decrease was 22 volumes per cent., while the least was 0.4 volumes per cent., and in the latter the operation lasted one and a half hours.

Comparing a series of ten patients to whom from 15 to 20 gm. of bicarbonate of sodium had been given intravenously before operation, with the same number who had not received such preliminary treatment, he found the decrease in the carbon dioxid combining capacity of the blood averaged 5.7 volumes per cent. as compared with an average of 9 in those who had not been given sodium bicarbonate.

He etherized two dogs for two hours or more, making carbon dioxid determinations every half hour. He found a reduction of from 10 to 12.5 volumes per cent., that is, from 22 to 24 per cent. of the normal amount. In one dog, the greatest reduction occurred in the third half hour; in the other the most rapid fall took place during the first half hour. Prolonged chloroform anesthesia in one dog caused a reduction of 24 volumes per cent., or 47 per cent. of the normal amount, which was considerably greater than that caused by ether.

Yandell Henderson [5] investigated the subject by experiments on dogs and reached the conclusion that the reduction in the carbon dioxid combining capacity of the blood is due to increased dissociation of the carbon dioxid from the blood, caused by the hyperpnea or increased respiratory activity produced by the irritant ether vapor. He contends that the decrease in the carbon dioxid content and capacity of the blood is due to increased ventilation of the lungs and not to actual reduction in the alkali reserve of the body.

Previous observations had been made by the Van Slyke apparatus with the plasma alone. Henderson used the whole blood and deter-

4. J. A. M. A. **68**:1391 (May 12) 1917.
5. J. Biol. Chem. **33**:345 (Feb.) 1918.

mined by means of his own apparatus the carbon dioxid content as well as its combining capacity when exposed to an atmosphere containing a uniform amount (5.5 per cent.) of carbon dioxid. He found the maximal decrease in the carbon dioxid content during ether anesthia was from 14 to 19 volumes per cent., while the greatest decrease in the carbon dioxid combining capacity was from 12 to 15 volumes per cent.

A state of hyperpnea was maintained by artificial respiration in dogs narcotized by chlorbutanol and he states that this produces as great a reduction in the carbon dioxid content and capacity of the blood as that seen in ether anesthesia, although the chlorbutanol alone does not produce that effect. He produced the opposite condition of decreased ventilation of the lungs by morphinizing dogs so as to reduce the respiratory activity, and he found an increase in the carbon dioxid content and capacity of the blood. When he produced light anesthesia with ether, so as to cause an acceleration of the respiration, he found a decrease in the carbon dioxid content and capacity of the blood; when he produced deep ether narcosis with feeble respiratory activity he found that both were increased.

In another series of experiments dogs inhaled ether vapor in an atmosphere containing from 6 to 7 per cent. of carbon dioxid, to which oxygen was added, and Henderson found there was no decrease in the carbon dioxid content or capacity of the blood, although the anesthesia was continued from three to five hours.

This last series of experiments is open to the criticism that the animals were compelled to breathe an artificial atmosphere containing a higher percentage of carbon dioxid than that of the alveolar air. Under such abnormal circumstances it is not surprising that there was an increase in the carbon dioxid of the blood.

EXPERIMENTS

Series 1.—The first series of experiments was made with blood plasma obtained by Van Slyke's method. The jugular vein was exposed after the animal became unconscious at the beginning of ether anesthesia and the first sample of blood was taken as the normal. Other samples were taken after half an hour, one hour, or one and a half hours of anesthesia and the carbon dioxid determinations were made with Van Slyke's apparatus.

Uniform anesthesia was maintained throughout each experiment by causing the animal to breathe ether vapor from a Mason jar of one liter capacity with an adjustable inlet tube and a respiratory valve in the top of the jar (Fig. 1), so that the vapor from the jar entered the cone with each inspiration, while the expired air escaped into the open without entering the jar. The jar was kept in a wooden box, the air of which was warmed by one or two eight-candle power

electric lamps, so that the air containing ether vapor was warmed before it entered the lungs.

The results are shown in Table 1. Only one out of twelve dogs did not show any decrease in the carbon dioxid combining capacity of the blood. Eleven showed a decrease varying from 5.6 to 23.3 volumes per cent., or from 15 to 46 per cent. of the normal. The average decrease at the end of the anesthesia was 10.8 volumes per

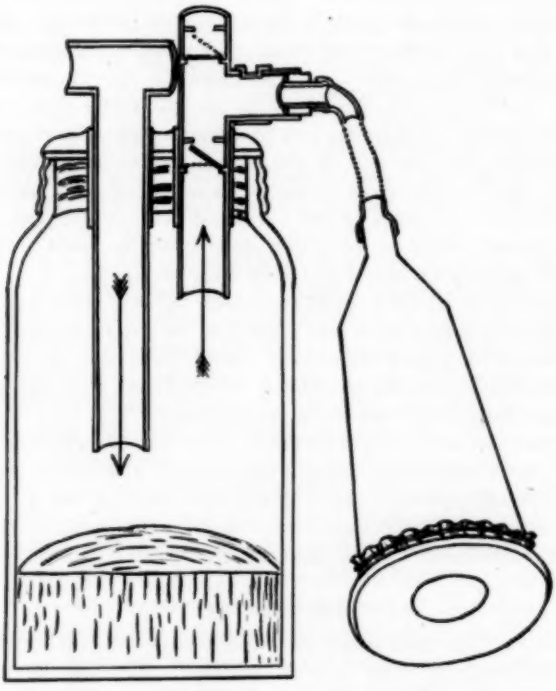

Fig. 1.—Sectional view of ether jar with respiratory valve on top. The continuous lines show the position of the light aluminum disks of the respiratory valve during inspiration, while the broken lines show the position of these disks during the phase of expiration.

cent. The anesthesia lasted one hour in three experiments and one and a half hours in nine experiments. Very little decrease occurred during the first half hour; the greatest decrease occurred during the second half hour, with a further fall during the third half hour period.

The same dog was used in Experiments 4, 5 and 6 on three consecutive days. There was no fall on the third day although there was a decrease on each of the first two days.

The same dog was used in Experiments 7, 8 and 9. The fall was no greater in Experiment 9, although a condition of shock was produced during the last two periods of that experiment by exposing and handling the abdominal viscera, so that the mean blood pressure was reduced from 140 mm. to less than 100 mm., and kept at that level.

One dog was also used for Experiments 11 and 12. This animal showed the greatest reduction in the alkali reserve on the first day, but less on the second day.

TABLE 1.—SUMMARY OF THE FIRST SERIES OF EXPERIMENTS. ORDINARY ETHER ANESTHESIA. UNIFORM ETHER VAPOR INSPIRED FROM ETHER JAR. EXPIRED AIR ESCAPED INTO THE OPEN WITHOUT ENTERING THE JAR. CARBON DIOXID DETERMINED BY VAN SLYKE'S METHOD

Experiment	Dog's Weight and Date of Experiment	Normal Carbon Dioxid Capacity of Blood	After ½ Hour of Anesthesia	After 1 Hour of Anesthesia	After 1½ Hours of Anesthesia	Decrease in Volume per Cent. (1-1½ Hrs.)	Decrease in Percentage of Normal
1	10.5 kg.	54.3	....	36.0	....	18.3	33.7
2	13 kg.	51.9	....	....	39.9	12.0	23.1
3	9.5 kg. Aug. 20.....	59.7	50.4	46.7	....	13.0	21.8
4	Same dog Aug. 22...	44.0	....	37.4	37.4	6.6	15.0
5	Same dog Aug. 23...	41.2	....	33.8	33.8	7.4	17.9
6	Same dog Aug. 24...	39.3	....	39.3	....	0.0	0.0
7	13 kg. Aug. 27, 10 a.m.	35.7	....	33.9	30.1	5.6	15.7
8	Same dog Aug. 27, 3 p.m.	33.9	....	26.4	24.6	9.3	27.4
9	Same dog Aug. 28...	42.1	43.0	41.2	34.7	7.4	17.5
10	7.7 kg.	52.05	42.2	37.5	39.3	12.7	24.4
11	5.0 kg. Sept. 5.....	49.7	43.2	31.05	26.4	23.3	46.8
12	Same dog Sept. 7....	59.9	54.3	54.3	46.1	13.8	23.0
	Average.....	46.98	46.6	37.95	34.7	10.8	22.1

Series 2.—In the second series of experiments, the normal blood was obtained before the anesthetic was started, by injecting procain and exposing the jugular vein without causing pain or struggling by the animal. Duplicate determinations were made from the normal blood to avoid error and these are given as the first and second normals.

In this series, and in all subsequent experiments throughout this investigation, the whole blood was used for determining the carbon dioxid content and the carbon dioxid combining capacity of the blood by Yandell Henderson's method and his apparatus was used in all experiments after the first series. The results by this method are more uniform and there is less danger of error than there is by Van Slyke's method.

The ether vapor was inhaled from a Mason jar of 1 liter capacity with the respiration valve in the top as in the first series. Anesthesia was maintained for two hours. Determinations of the carbon dioxid content and carbon dioxid combining capacity of the blood were made at the end of one and two hours of anesthesia, respectively, and also half an hour and one hour after the end of the anesthesia, to determine the after-effect.

The details of these experiments are not given for each separate experiment but they may be summarized by giving the averages of the different periods in the five experiments of this series (Table 2).

All of the experiments of this series showed a decrease, varying from 4.0 to 8.6 volumes per cent. in the carbon dioxid combining capacity of the blood. The results were quite uniform and did not show as much variation as in the first series.

One important fact brought out by this series is that there was practically no decrease in the alkali reserve at the end of one hour of etherization, but the entire reduction occurred during the second hour. This has a practical bearing as it shows that the danger of decrease in the alkali reserve is greater from prolonged anesthesia than it is when this is not continued for more than an hour.

TABLE 2.—Summary of Five Experiments. Ordinary Ether Anesthesia. Carbon Determinations by Henderson's Method

Periods for Which Averages Are Given in Last Two Columns	Average Carbon Dioxid Content of Blood	Average Carbon Dioxid Combining Capacity of Blood
First normal (blood obtained by procain)	48.27	48.77
Second normal (blood obtained by procain)	48.04	49.02
After 1 hour of ordinary ether anesthesia	48.45	48.28
After 2 hours of ordinary ether anesthesia	42.86	42.71
One-half hour after end of anesthesia	42.70	43.28
One hour after end of anesthesia	47.09	46.79
Average decrease in volume per cent.	5.28	6.00
Average decrease in percentage of normal	.....	12.5

The reduction in the carbon dioxid content and combining capacity of the blood continued for half an hour after the end of anesthesia in each experiment but both had practically returned to the normal at the end of one hour.

Series 3.—A third series of experiments was made to determine whether there is an actual reduction in the alkali reserve from ether anesthesia, or if this is an apparent condition caused by the hyperpnea or increased respiratory activity as a result of the irritant effects of ether vapor, as claimed by Henderson. The normal blood was obtained from the vein after it had been exposed by the use of procain. The animal was first etherized in the ordinary way until tracheotomy could be performed and etherization was then continued by connecting the tracheal tube to a metal tube in the top of a 2-quart Mason jar containing ether. A large rubber tube from the Hans Meyer artificial respiration apparatus was connected to another tube in the top of the ether jar. This apparatus can be adjusted to force through the ether jar and into the lungs the same amount of air that is breathed in natural respiration by animals of different size. The tidal air had been determined by means of a spirometer for each animal before

the experiment started, and the artificial respiration apparatus was adjusted to furnish the same amount. It also aspirates from the jar and lungs in expiration the same volume of air that is forced in under positive pressure during inspiration. Of course the air in the jar, 2 liters, was added to the dead air space of the upper respiratory passages and analysis of this air in each experiment showed that it contained on the average from 2.6 to 3.0 per cent. of carbon dioxid. The animals used in the third series were, therefore, furnished with ether in a uniform vapor density, in an atmosphere containing approximately 3 per cent. of carbon dioxid. This accounts for the increase in the carbon dioxid content and combining capacity of the blood which occurred after one hour of anesthesia maintained in this way. The increased carbon dioxid of the air would tend to keep up the carbon dioxid of the blood, even if there were a condition of hyperpnea. Hyperpnea was prevented in this series by the artificial respiration which maintained a uniform respiratory rate and volume throughout the entire experiment.

TABLE 3.—SUMMARY OF THREE EXPERIMENTS OF SERIES III. EFFECT OF ETHER ANESTHESIA BY ARTIFICIAL RESPIRATION TO MAINTAIN A UNIFORM RESPIRATORY RATE AND VOLUME, THUS ELIMINATING HYPERPNEA

Periods for Which Averages Are Given	Carbon Dioxid Content of Blood	Carbon Dioxid Combining Capacity of Blood
First normal (blood obtained by use of procain)	51.82	54.18
Second normal (blood obtained by use of procain)	51.95	53.02
After one hour of anesthesia by artificial respiration through a two liter jar containing 2.6-3.0 per cent. carbon dioxid	53.02	56.15
After two hours of anesthesia	46.06	50.06
After three hours of anesthesia	42.12	44.75
Average decrease in volume per cent. after two hours etherization	5.82	3.54
Average decrease in volume per cent. after three hours etherization	9.76	8.85
Maximal decrease in percentage of the normal amount	18%	16.5%

The elimination of hyperpnea in the experiments of the third series shows that this condition is not responsible for the reduction in the alkali reserve in the experiments of the first and second series where ordinary ether anesthesia was used. In spite of the increased percentage of carbon dioxid in the tidal and stationary or alveolar air, which in itself caused an increase in the carbon dioxid content and combining capacity of the blood during the first hour of anesthesia, there was a decided fall during the second hour and a further fall during the third hour. The fall during the second hour was less than that in the second series, but when the increase during the first hour is taken into consideration, the results are practically the same in both series. The reduction for three hours of anesthesia by artificial respiration is greater than that in the second series and almost as great as the average of the experiments in the first series.

The details of each experiment are not given on account of limited space. The results were quite uniform in the three experiments and these are summarized in Table 3 by giving the averages for the different periods.

Series 4.—This series of experiments was made to determine whether the ventilation of the lungs is increased in proportion to the acceleration of the respiratory rate during ordinary anesthesia; also to devise a method of overcoming the reduction in the carbon dioxid content and capacity of the blood that might be produced by a simple hyperpnea, or increased in the volume of air moved in and out of the lungs, occurring with natural respiration.

The experiments were divided into four periods of half an hour each. The respiratory volume, as measured by a spirometer, and the rate were taken for two or three minutes and averaged for one minute periods. At least six observations were made in each period at intervals of from three to five minutes and the average was taken for the entire period of half an hour.

During the first period the animal breathed through the cone and Mason jar with the respiration valve in the top, but without ether in the jar. The expired air escaping from the top of the respiration valve was collected and measured in a spirometer. This period was taken as the normal for comparison with the other periods.

During the second period the dog breathed through a jar with a respiration valve on the far side from the animal (Fig. 2). In this way the inspired and expired air passed through the jar, the contents of which became part of the dead air space of the cone and upper respiratory passages. Analysis of the air of the jar showed that it contained from 16 to 17 per cent. of oxygen and from 2.5 to 3.0 per cent. of carbon dioxid. Jars of different size (1, 2 and 3 liters) were used, but the composition of the air did not vary much. When such jars are used for vaporizing ether the large size (3 liters) is desirable as the vapor density of the ether does not then exceed the amount desirable for anesthesia.

The third period was like the first, except that ether was placed in the jar for ordinary ether anesthesia.

The fourth period was like the second, except that ether, in sufficient amount to keep the bottom covered, was placed in the jar through which the inspired and expired air passed, so that the animal inhaled ether vapor in an atmosphere containing from 16 to 17 per cent. of oxygen and from 2.5 to 3 per cent. of carbon dioxid.

Six experiments were made in this series and the averages for the different periods are given in Table 4, together with the percentage for each period over the normal. It will be seen that an atmosphere containing from 2.5 to 3 per cent. of carbon dioxid (Period B)

increased the respiratory volume 74 per cent., but only increased the rate 7 per cent. The greater depth of the respirations is caused by the chemical stimulation of the respiratory center produced by the increased tension of carbon dioxid in the blood.

Ether vapor alone in the ordinary atmosphere (Period C) increased the respiratory rate 107 per cent., but only increased the respiratory volume 33 per cent. This indicates that the acceleration of the respiratory rate in ordinary ether anesthesia is three times greater than the increase in the ventilation of the lungs, and that the hyperpnea is not so great as it appears to be when the number of shallow respirations alone are considered.

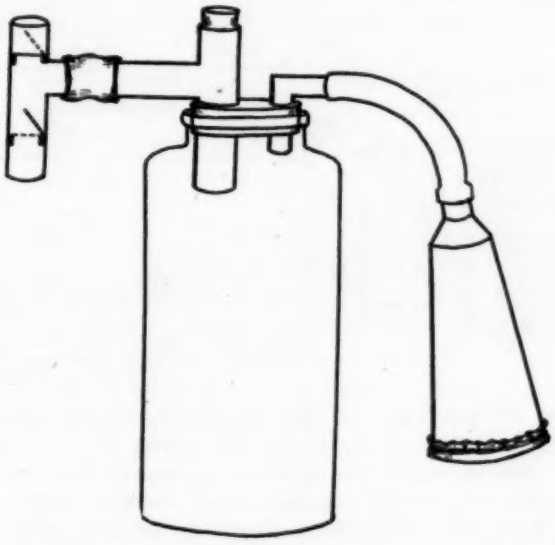

Fig. 2.—Ether jar with respiratory valve on the far side from the cone. The continuous lines show the position of the disks of the valve during inspiration, while the broken lines show the position during expiration.

The average for the fourth period (Period D) showed that the ether vapor in an atmosphere containing 3 per cent. of carbon dioxid caused an increase of 80 per cent. in the rate but the volume is increased less than 5 per cent. The latter varies with the depth of anesthesia, but the increase in the volume is never as great as with ether alone, or an atmosphere of 3 per cent. of carbon dioxid alone. Apparently the depression of the respiratory center by the ether lessens its irritability to chemical stimulation by an increased tension of carbon dioxid

in the blood. The method used in this last period makes it possible to study the effect of etherization on the alkali reserve with natural respiration when there is comparatively little increase in the ventilation of the lungs. Any increase in the volume of air is fully offset by the increased percentage of carbon dioxid in the alveolar air. This was shown by the increased amount of carbon dioxid in the blood which was found in numerous experiments.

TABLE 4.—SUMMARY OF SIX EXPERIMENTS TO DETERMINE THE EFFECT OF (1) ORDINARY ETHER ANESTHESIA ON RESPIRATORY RATE AND VOLUME; (2) BREATHING THROUGH A JAR CONTAINING 16-17% OXYGEN AND 2.5-3% CARBON DIOXID; (3) ETHER VAPOR IN A JAR CONTAINING 2.5-3% CARBON DIOXID. SIX OBSERVATIONS MADE FOR EACH PERIOD AT INTERVALS OF THREE TO FIVE MINUTES

Half-hour Periods for Which Averages Were Made	Average Respiratory Rate per Minute	Percentage Increase Over Normal	Average Respiratory Volume (Cubic Inches per Minute)	Percentage Increase Over Normal
A. Normal respiration through Mason jar with respiration valve in top, i. e., breathing ordinary atmosphere	26	...	100	
B. Breathing through jar with respiration valve on far side. "Dead air space" in jar contained 16-17% oxygen and 2.5-3.0% carbon dioxid	28	7	190	74
C. Ether anesthesia. Uniform ether vapor from jar with valve in top, i. e., expired air escaped without entering jar	54	107	145	33
D. Ether anesthesia through jar with valve on far side, i. e., ether vapor in air containing 16-17% oxygen and 2.5-3.0% carbon dioxid	47	80.7	113	3.7

It was not practicable to make simultaneous observations of the respiratory rate and volume and of the carbon dioxid of the blood in many experiments, but the following experiment shows no decrease in the carbon dioxid combining capacity of the blood during one and one half hours of ordinary ether anesthesia, although there was considerable hyperpnea. During the second period of one and one-half hours in the same animal, when the animal inhaled ether vapor in air containing from 3 to 3.5 per cent. of carbon dioxid, there was a decided decrease in the alkali reserve of the blood, although the respiratory volume was practically the same as it had been during the first period.

Series 5.—The experiments of this series were made to determine the effect on the carbon dioxid content and combining capacity of the blood produced (1) by natural respiration through a 3 liter jar containing approximately 3 per cent. of carbon dioxid, and (2) by inhaling ether vapor from such a jar for two or three hours. Observations were also made one and one-half hours after the end of the anesthesia

when it was found that the alkali reserve of the blood had practically returned to normal.

Six experiments were made in this series which are summarized in Table 6 by giving the averages for each period. The dogs breathed through a 3 liter bottle with the respiratory valve on the distal side from the animal and described in discussing results in Period D of Series 4. The air of the bottle was vitiated by respiration so that it contained from 16 to 17 per cent. of oxygen and 3 per cent. of carbon dioxid. The bottom of the bottle was kept covered with ether.

TABLE 5.—EXPERIMENT 12. SHOWING SIMULTANEOUS OBSERVATIONS ON RESPIRATORY RATE AND VOLUME AND CARBON DIOXID CONTENT AND CAPACITY OF THE BLOOD (A) DURING ORDINARY ETHER ANESTHESIA AND (B) DURING ETHER ANESTHESIA WHILE BREATHING 3.5% CARBON DIOXID

	Respiratory Volume (Cubic Inches per Minute)	Respiratory Rate per Minute	Carbon Dioxid Content of Blood	Combining Capacity of Blood
9:30 a. m. First normal (blood taken by procain)	...	...	55.72	57.30
Second normal	...	...	53.80	58.70
Average	...	...	54.76	58.00
9:40-9:50 Average of 4 observations	90	31		
10.00 Started ether anesthesia				
10:00-10:25 Average of 8 observations	186	41		
10:30	...	...	50.90	60.70
10:55-11:25 Average of 7 observations	212	54		
11:30 After 1½ hours of ordinary ether anesthesia	...	...	59.20	63.00
11:32 Began inhalation of ether vapor with 3.5% carbon dioxid by breathing through a 3 liter jar				
11:40-11:55 Average of 4 observations	242	83		
12:00 m.	...	...	46.70	49.94
12:45-12:55 Average of 3 observations	210	86		
1:00 p. m. After 3 hours of anesthesia, i. e., 1½ hours with ordinary air and 1½ hours of ether vapor with 3-3.5 carbon dioxid	...	...	45.92	47.94
Decrease	...	...	8.84	10.06

In the experiments of Series 3, artificial respiration was used through a 2 liter ether jar; in those of Series 5, natural respiration was used through a 3 liter ether jar. In both series the jar contained approximately 3 per cent. of carbon dioxid. Hyperpnea was completely eliminated in Series 3, but not in Series 5. Even if there were any hyperpnea in Series 5, it did not cause any increased dissociation of carbon dioxid from the blood on account of the increased percentage of carbon dioxid in the tidal and in the alveolar air. This is clearly shown by the increase in the carbon dioxid content and combining capacity of the blood when the animal breathed through the jar for half an hour before the anesthetic was started. The animals breathed

quietly during this time and did not struggle. Under such conditions any decrease in the carbon dioxid combining capacity of the blood that occurred in the six experiments of Series 5 could not have been due to hyperpnea.

The reduction was practically the same as that in Series 2, in which the animals inhaled ether vapor in the ordinary atmosphere, and it was three-fourths as great (6.4 volumes per cent.) as that in Series 3 (8.85 volumes per cent.).

TABLE 6.—SERIES 5. SUMMARY OF SIX EXPERIMENTS WITH NATURAL RESPIRATION. ETHER VAPORIZED IN A THREE-LITER JAR CONTAINING 3 PER CENT CARBON DIOXID

Periods for Which Averages Are Given in Last Two Columns	Average Carbon Dioxid Content of Blood	Average Carbon Dioxid Combining Capacity of the Blood
First normal (vein exposed by procain)	52.1	52.3
Second normal	53.2	54.1
After breathing through 3 liter bottle containing 3% carbon dioxid (without ether) for one-half hour	56.3	57.6
After one hour of etherization (ether vaporized in jar containing 3% carbon dioxid through which the animal breathed)	54.6	54.6
After two hours of anesthesia (6 experiments)	48.5	49.4
After three hours of anesthesia (3 experiments)	46.1	45.4
Average decrease in volumes per cent.	6.16	6.4
Average decrease in percentage of the normal amount	11.1%	11.6%
One and one-half hours after end of anesthesia (4 exper.)	52.7	52.1

As an atmosphere containing 3 per cent. of carbon dioxid causes a distinct increase of 3 or 4 volumes per cent. in the carbon dioxid content and combining capacity of the blood, this method seems much better than the one used by Henderson, in which the animals breathed an atmosphere containing 6 or 7 per cent. of carbon dioxid with the addition of oxygen.

That hyperpnea is not a factor in the decrease in the alkali reserve produced by ether anesthesia is also evident from the fact that the decrease does not occur during the first hour of anesthesia, when the greatest acceleration of the respiratory rate occurs, but during the second or third hour when the respirations are uniform in rate. It is also evident from the six experiments of Series 4 that the increase in respiratory volume is only about one-third as great as the increase in rate in the hyperpnea which occurs early in ether anesthesia.

THE EFFECT OF DECREASED OXYGEN

It is a well known fact that a decreased supply of oxygen to the tissues may produce a decrease in the alkali reserve. In order to determine whether the decrease in the oxygen in the 3 liter jar, through which the animal breathed in order to increase the carbon

dioxid of the tidal and alveolar air, was responsible for the decrease in the carbon dioxid combining capacity of the blood during ether anesthesia by this method, three control experiments were made.

The dogs were kept quiet by giving chlorbutanol to produce narcosis after the normal sample of blood had been taken by exposing the vein with the use of procain. They were then made to breathe through the jar of 3 liters capacity for three hours and the carbon dioxid content and capacity of the blood were determined as before. Analysis of the air in the jar showed an average of 16.3 per cent. oxygen and 3.9 per cent. carbon dioxid. The averages of the three experiments are shown in Table 7.

TABLE 7.—Effect of Decreased Oxygen

	Carbon Dioxid Content	Carbon Dioxid Combining Capacity
Normal blood	58.5	62.6
After one-half hour of chlorbutanol narcosis	48.7	55.7
After breathing through 3 liter jar for 1 hour	59.2	60.0
After breathing through 3 liter jar for 2 hours	57.0	58.4
After breathing through 3 liter jar for 3 hours	60.1	58.6

These experiments show conclusively that the decrease in the oxygen in the air breathed from the jar was not responsible for the decrease in the alkali reserve of the blood. The chlorbutanol narcosis seemed to cause a distinct decrease until the respiratory center was stimulated by the increased tension of carbon dioxid in the blood which resulted from a higher percentage of carbon dioxid in the alveolar air. Henderson states that chlorbutanol does not influence the carbon dioxid combining capacity of the blood. However, it lessens the respiratory activity decidedly.

AFTER-EFFECT OF ETHER ANESTHESIA

All five of the experiments of the second series showed that the reduction in the alkali reserve at the end of etherization for two hours continued for half an hour after the anesthesia ended, but the carbon dioxid content and capacity returned almost to the normal at the end of one hour after the anesthesia.

Four of the experiments in Series 5 showed that the carbon dioxid content and capacity of the blood had returned to the normal one and a half hours after the termination of the anesthesia, which lasted for two hours in two experiments, and for three hours in two experiments.

Two other experiments were made to determine if there is a secondary fall. There was an average reduction of 8.5 volumes per cent. in the carbon dioxid combining capacity at the end of two hours

of ordinary anesthesia and this continued for one hour. At the end of two and one half hours the alkali reserve had returned to normal and remained so at the end of four and five hours, respectively, after the anesthesia.

CONCLUSIONS

1. Ordinary ether anesthesia, without any of the contributing conditions that attend surgical operations, causes a distinct decrease in the alkali reserve. The decrease in carbon dioxid combining capacity of the blood of dogs is usually from 6 to 10 volumes per cent.

2. There is comparatively little diminution during the first hour but it occurs almost entirely after that time and is in direct proportion to the duration of the anesthetic.

3. There is an actual decrease in the alkali reserve and not an apparent condition due to hyperpnea. The latter is most marked early in the anesthesia but there is little or no dealkalization during the first hour. The usual decrease occurs when anesthesia is maintained by artificial respiration which provides a uniform respiratory volume; also when an animal breathes an atmosphere containing 3 per cent. of carbon dioxid in which ether has been vaporized.

4. Breathing an atmosphere containing 16 per cent. of oxygen and 3.5 per cent. of carbon dioxid for three hours does not diminish the alkali reserve.

5. The greatest decrease in the alkali reserve produced by ether anesthesia occurs at the end of the anesthesia and remains at that level for from one-half to one hour after the anesthesia, at a time when there is decreased respiratory activity. Following this brief after-effect, there is a rapid increase in the alkali reserve and it returns to the normal in from one to two hours after the anesthesia.

All of the experiments presented in this paper were performed on normal dogs. The decrease in the alkali reserve never reached a dangerous level and it only continued for a short time after the anesthesia. It is impossible to conclude from them what might occur if there is a reduction from altered metabolism before a surgical operation, or when a patient is in a condition of surgical shock which is attended by a reduction in the carbon dioxid combining capacity of the blood. Such conditions, added to that produced by ether, may be more serious. It should also be remembered that it is extremely difficult to produce in dogs the condition known as acidosis, even by injecting large amounts of a mineral acid into the circulation, as they are able to protect themselves against acids by the alkali reserve of the body and by the ability to form ammonia salts from protein metabolism.

STUDIES ON BLOOD SUGAR—EFFECT OF BLOOD CONSTITUENTS ON PICRATE SOLUTIONS

A CONSIDERATION OF THE LIMITATIONS OF THE MODIFIED LEWIS-BENEDICT TEST *

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While working on a blood sugar problem we became interested in the statement of McLean [1] that "it is doubtful whether the picric acid method is at all reliable in the case of whole blood on account of the creatinin (and probably other interfering substances)" which under the conditions of the estimation gives a color similar to that produced by glucose in the presence of picric acid. De Wesselow [2] claims to show that the Benedict method with whole blood gives 45 per cent. higher readings than the McLean method which utilizes an entirely different principle—precipitation of protein with heat and dialyzed iron and a procedure which is thought to overcome the tendency to oxidation of cuprous oxid during titration.

We have done so much work with the Lewis-Benedict method, and it has been so satisfactory in our hands as a clinical test, that we decided to satisfy ourselves as to the correctness of these statements before giving it up as the routine method in the clinic.

The results of our investigation may be of interest. We have done the following experiments which may have some bearing on the reliability of the Lewis-Benedict test and which may give some added information concerning substances which appear in the blood under abnormal conditions that may influence blood sugar records and explain the occurrence of "hyperglycemia" without glycosuria.

The Effect of Oxalation.—Blood was drawn from four diabetics and one normal person. As quickly as the blood was drawn, 2 c.c. were placed in a volumetric flask, laked with distilled water and precipitated in the ordinary way with picrate solution. The blood was not oxalated. The analysis was carried through immediately. The remainder of the blood was oxalated and determinations were made by the Lewis-Benedict method one half hour later (Table 1).

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* From the Department of Pediatrics and Infectious Diseases, University of Michigan Hospital.

* Read before the American Pediatric Society, May 30, June 1-2, 1920.

1. Maclean, H.: On the Estimation of Sugar in Blood, Biochem. Jr. **13**:135, 1919.

2. De Wesselow, C. L. V.: Picric Acid Method for Estimation of Sugar in the Blood and Comparison of This Method with that of Maclean, Biochem. Jr. **13**:148, 1919.

This shows that oxalation produces no difference in the reaction.

The Effect of Creatinin.—We have shown by a simple gross experiment that creatinin added directly to a 2 c.c. sample of blood increases the blood sugar record given by the Lewis-Benedict method (Table 2).

These amounts of creatinin are many times greater than, probably, ever occur in blood in disease. The highest blood creatinin we find recorded is 33.3 mg. per cent. in a case of nephritis due to mercurial poisoning reported by Meyers and Killian.[3]

We next attempted to determine the lowest limit of sensitivity of the picrate solution to creatinin in water solutions and in blood (Tables 3 and 4).

TABLE 1.—EXPERIMENT 1. Nov. 12, 1919

Case	Nonoxalated Sample	Oxalated Sample
Labar	0.204	0.200
Cassel	0.175	0.170
Germain	0.300	0.300
Fregel	0.312	0.312
Kozolowski (control)	0.117	0.120

TABLE 2.—EXPERIMENT 2. Nov. 22, 1919

Control Blood	2 c.c. Blood + 1 mg. Creatinin = 50 mg. per 100	2 c.c. Blood + 2 mg. Creatinin = 100 mg. per 100
0.125%	0.180%	0.200%

In Series 2 of this experiment a distinct color change with a 2 c.c. sample of a creatinin solution containing 2.5 mg. in 100 c.c. of water is shown. In other words, the picrate solution is sensitive to 0.00005 gm. creatinin in water.

A distinct color change occurs in the picrate solution when 2 c.c. of a solution of 6 or 7 mg. of creatinin to 100 c.c. of water are added to a 2 c.c. sample of normal blood (Table 5). In other words, in order that creatinin in the blood may have any influence on a Lewis-Benedict blood sugar record, it must be increased at least 6 or 7 mg. above normal which would give a creatinin value of 9 mg. per cent. or above.

The remarkable sensitiveness of the picrate solution is better comprehended when we recognize the fact that in the 2 c.c. sample of blood, plus the creatinin solution, there is only 0.00016 gm. of creatinin present.

3. Meyers, V. C., and Killian, J. A.: Prognostic Value of Creatinin of the Blood in Nephritis, Am. J. M. Sc. **157**:674, 1919.

We have seen that the highest blood creatinin is 33.3 mg. per cent.[3] If the record obtained by this amount (0.045, Table 5) added to blood is added to a normal blood sugar record, $0.150 + 0.45 = 0.195$, the upper border of normal is certainly reached. If, on the other hand, a case of diabetes was complicated by a marked renal disturbance inducing a high blood creatinin, one might expect a considerable increase in the blood sugar record not due to glucose.

TABLE 3.—EXPERIMENT 3. DEC. 29, 1919. CREATININ IN WATER. SERIES 1

Size of Water Sample	Creatinin		Blood Sugar Record, L.-B. Test
	Amount Added	Strength of Solution	
2 c.c. control	.....	.....	0.040
2 c.c.	2 c.c.	200 mg. in 100 c.c. water	0.333
2 c.c.	2 c.c.	190 mg. in 100 c.c. water	0.285
2 c.c.	2 c.c.	180 mg. in 100 c.c. water	0.270
2 c.c.	2 c.c.	170 mg. in 100 c.c. water	0.270
2 c.c.	2 c.c.	160 mg. in 100 c.c. water	0.270
2 c.c.	2 c.c.	150 mg. in 100 c.c. water	0.250
2 c.c.	2 c.c.	140 mg. in 100 c.c. water	0.250
2 c.c.	2 c.c.	130 mg. in 100 c.c. water	0.222
2 c.c.	2 c.c.	120 mg. in 100 c.c. water	0.225
2 c.c.	2 c.c.	110 mg. in 100 c.c. water	0.222
2 c.c.	2 c.c.	100 mg. in 100 c.c. water	0.222
2 c.c.	2 c.c.	90 mg. in 100 c.c. water	0.215
2 c.c.	2 c.c.	80 mg. in 100 c.c. water	0.220
2 c.c.	2 c.c.	70 mg. in 100 c.c. water	0.220
2 c.c.	2 c.c.	60 mg. in 100 c.c. water	0.182
2 c.c.	2 c.c.	50 mg. in 100 c.c. water	0.166
2 c.c.	2 c.c.	40 mg. in 100 c.c. water	0.143
2 c.c.	2 c.c.	30 mg. in 100 c.c. water	0.135
2 c.c.	2 c.c.	20 mg. in 100 c.c. water	0.090
2 c.c.	2 c.c.	10 mg. in 100 c.c. water	0.090
2 c.c. control	.....	.....	0.040

TABLE 4.—EXPERIMENT 3. DEC. 29, 1919. CREATININ IN WATER. SERIES 2

Size of Water Sample	Creatinin		Blood Sugar Record, L.-B. Test
	Amount Added	Strength of Solution	
2 c.c. control	.....	.....	0.040
2 c.c.	2 c.c.	60 mg. in 100 c.c. water	0.200
2 c.c.	2 c.c.	50 mg. in 100 c.c. water	0.160
2 c.c.	2 c.c.	40 mg. in 100 c.c. water	0.133
2 c.c.	2 c.c.	30 mg. in 100 c.c. water	0.111
2 c.c.	2 c.c.	20 mg. in 100 c.c. water	0.080
2 c.c.	2 c.c.	10 mg. in 100 c.c. water	0.070
2 c.c.	2 c.c.	5 mg. in 100 c.c. water	0.050
2 c.c.	2 c.c.	2½ mg. in 100 c.c. water	0.050
2 c.c. control	.....	.....	0.040

The Effect of Urea.—If 4 or 5 mg. urea crystals are added to a 2 c.c. sample of blood, a color change in the picrate solution may be obtained. This amount is far above the amount that ever occurs in the circulating blood. With graded and accurately measured amounts of urea, such as might occur as excess in circulating blood, no change in color whatsoever takes place (Tables 6 and 7).

Dilutions of urea in blood exactly like those tabulated for water were set up and run. There was absolutely no change produced by these amounts of urea.

TABLE 5.—Experiment 4. Jan. 2, 1920. Creatinin in Blood. Series 1

Size of Blood Sample	Creatinin		Blood Sugar Record, L.-B. Test	Difference from Control Average
	Amount Added	Strength of Solution		
2 c.c.*	.....	.....	0.118	
2 c.c.	2 c.c.	190 mg. in 100 c.c. water	0.475	0.356
2 c.c.	2 c.c.	180 mg. in 100 c.c. water	0.330	0.211
2 c.c.	2 c.c.	170 mg. in 100 c.c. water	0.330	0.221
2 c.c.	2 c.c.	160 mg. in 100 c.c. water	0.320	0.201
2 c.c.	2 c.c.	150 mg. in 100 c.c. water	0.350	0.131
2 c.c.	2 c.c.	140 mg. in 100 c.c. water	0.222	0.108
2 c.c.	2 c.c.	130 mg. in 100 c.c. water	0.222	0.103
2 c.c.	2 c.c.	120 mg. in 100 c.c. water	0.224	0.105
2 c.c.	2 c.c.	110 mg. in 100 c.c. water	0.308	0.189
2 c.c.	2 c.c.	100 mg. in 100 c.c. water	0.247	0.128
2 c.c.	2 c.c.	90 mg. in 100 c.c. water	0.247	0.128
2 c.c.	2 c.c.	80 mg. in 100 c.c. water	0.243	0.124
2 c.c.	2 c.c.	70 mg. in 100 c.c. water	0.218	0.099
2 c.c.	2 c.c.	60 mg. in 100 c.c. water	0.221	0.102
2 c.c.	2 c.c.	50 mg. in 100 c.c. water	0.198	0.079
2 c.c.	2 c.c.	40 mg. in 100 c.c. water	0.165	0.046
2 c.c.	2 c.c.	30 mg. in 100 c.c. water	0.164	0.045
2 c.c.	2 c.c.	20 mg. in 100 c.c. water	0.140	0.021
2 c.c.	2 c.c.	10 mg. in 100 c.c. water	0.132	0.013
2 c.c.*	.....	.....	0.120	
Series 2				
2 c.c.*	.....	.....	0.110	
2 c.c.	2 c.c.	10 mg. in 100 c.c. water	0.141	0.030
2 c.c.	2 c.c.	9 mg. in 100 c.c. water	0.143	0.033
2 c.c.	2 c.c.	8 mg. in 100 c.c. water	0.127	0.016
2 c.c.	2 c.c.	7 mg. in 100 c.c. water	0.122	0.011
2 c.c.	2 c.c.	6 mg. in 100 c.c. water	0.118	0.007
2 c.c.	2 c.c.	5 mg. in 100 c.c. water	0.117	0.006
2 c.c.	2 c.c.	4 mg. in 100 c.c. water	0.117	0.006
2 c.c.	2 c.c.	3 mg. in 100 c.c. water	0.118	0.007
2 c.c.	2 c.c.	2 mg. in 100 c.c. water	0.111	
2 c.c.	2 c.c.	1 mg. in 100 c.c. water	0.108	
2 c.c.*	.....	.....	0.113	

* Control.

TABLE 6.—Experiment 5. May 20, 1920. Urea in Water

Size of Water Sample	Urea		Blood Sugar Record, L.-B. Test
	Amount Added	Strength of Solution	
2 c.c. control	.....	.....	0.040
2 c.c.	2 c.c.	300 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	200 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	100 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	90 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	80 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	60 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	50 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	40 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	30 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	20 mg. in 100 c.c. water	0.040
2 c.c.	2 c.c.	10 mg. in 100 c.c. water	0.040

TABLE 7.—Experiment 6, May 20, 1920. Urea in Blood

Size of Blood Sample	Urea		Blood Sugar Record, L.-B. Test
	Amount Added	Strength of Solution	
2 c.c. control	.....	.....	0.112
2 c.c.	2 c.c.	300 mg. in 100 c.c. water	0.113

From the above experiment it will be seen that in these amounts urea does not interfere.

Uric acid does not interfere. Ammonia nitrogen does not interfere.

The Effect of the Amino Acids.—Glycocol: Two c.c. of water plus a knife point of glycocol. Boiling for ten minutes produces no change. On standing twenty minutes no change. After complete cooling one hour and fifteen minutes, consolidation in both tubes, the glycocol tube shows a definite change. After reheating for ten minutes the readings shown in Table 9 were obtained:

TABLE 8.—EXPERIMENT 7. MAY 22, 1920. THE EFFECT OF URIC ACID

2 c.c. water control	0.040
2 c.c. water + 2 mg. uric acid	0.040
2 c.c. blood control	0.100
2 c.c. blood + 1 mg. uric acid	0.100

TABLE 9.—EXPERIMENT 8. EFFECT OF GLYCOCOL

2 c.c. water control	0.040
2 c.c. glycocol tube	0.190

TABLE 10.—EXPERIMENT 9. EPINEPHRIN IN WATER

Size of Water Sample	Epinephrin		Blood Sugar Record	Water Control Record	Date and Remarks
	Amount Added	Strength of Solution			
2 c.c.	1.5 c.c.	1:1000 water	0.800	0.040	Dec. 5, 1919, open bottle epinephrin
2 c.c.	2.0 c.c.	1:1000 water	1.000		
2 c.c.	2.0 c.c.	1:1000 water	0.900	0.040	Dec. 8, 1919, new bottle epinephrin
2 c.c.	1.0 c.c.	1:1000 water	0.450		
2 c.c.	7 minims	1:1000 water	0.200		
2 c.c.	3 minims	1:1000 water	0.115		
2 c.c.	1 minim	1:1000 water	0.070		
2 c.c.	2.0 c.c.	1:1000 water	0.337	.....	March 27, 1920, epinephrin had stood in corked bottle since Dec. 8, 1919
2 c.c.	2.0 c.c.	1:2000 water	0.205		
2 c.c.	2.0 c.c.	1:4000 water	0.148		
2 c.c.	2.0 c.c.	1:5000 water	0.125	.....	New bottle May 25, 1920
2 c.c.	2.0 c.c.	1:7500 water	0.111		
2 c.c.	2.0 c.c.	1:10,000 water	0.095		
2 c.c.	2.0 c.c.	1:20,000 water	0.075		
2 c.c.	2.0 c.c.	1:50,000 water	0.060		
2 c.c.	2.0 c.c.	1:100,000 water	0.040		
2 c.c.	2.0 c.c.	1:200,000 water	0.040		
2 c.c.	2.0 c.c.	1:400,000 water	0.040		
2 c.c.	2.0 c.c.	1:800,000 water	0.040		

Tyrosin, taurin, tryptophan and alanin induce no change. These are all the amino acids we could find.

The Effect of Epinephrin.—If a drop or two of epinephrrin solution (1:1,000) is added to picrate, Fehling's or Benedict's solution, a distinct reduction takes place. The experiments tabulated in Table 10 show the lowest limit of sensitivity of the picrate solution to epinephrin in water and in blood.

The lowest limit of sensitivity in water is 2 c.c. of a 1:50,000 solution, or 0.00004 gm. The difference in the records obtained with the same dilution is explained by the condition of the sample, age, etc.

The experiment tabulated in Table 11 shows that epinephrin in infinitesimally small amounts induces a color change when added to blood. The lowest limit of sensitivity of the picrate solution of the Lewis-Benedict test to epinephrin added to blood is safely placed at 0.000025 per cent., or in other words, 0.025 mg. per cent. If we calculate the amount present in a 2 c.c. sample of blood plus 2 c.c. of a 1:4,000,000 solution, the picrate solution reacts to 0.0000005 gm. of epinephrin (adrenalin). From this it will be seen that the picrate solution is many-fold more sensitive to solutions of epinephrin added to blood than to water.

TABLE 11.—EXPERIMENT 10. APRIL 30, 1920. EPINEPHRIN IN BLOOD

Size of Water Sample	Epinephrin		Blood Sugar Record, L.-B. Test
	Amount Added	Strength of Solution	
2 c.c. control	.....	.....	0.127
2 c.c. control	.....	.....	0.120
2 c.c.	2 c.c.	1 mg. in 100,000 c.c. water	0.196
2 c.c.	2 c.c.	1 mg. in 200,000 c.c. water	0.171
2 c.c.	2 c.c.	1 mg. in 400 000 c.c. water	0.165
2 c.c.	2 c.c.	1 mg. in 500,000 c.c. water	0.153
2 c.c.	2 c.c.	1 mg. in 1,000,000 c.c. water	0.140
2 c.c.	2 c.c.	1 mg. in 1,500,000 c.c. water	0.138
2 c.c.	2 c.c.	1 mg. in 2,000,000 c.c. water	0.136
2 c.c.	2 c.c.	1 mg. in 2,500,000 c.c. water	0.135
2 c.c.	2 c.c.	1 mg. in 3,000,000 c.c. water	0.133
2 c.c.	2 c.c.	1 mg. in 4,000,000 c.c. water	0.130
2 c.c.	2 c.c.	1 mg. in 2,000 000 c.c. water	0.160
2 c.c.	2 c.c.	1 mg. in 1 000,000 c.c. water	0.176
2 c.c. control	.....	.....	0.114

THE ACETONE BODIES

The Effect of Acetone.—Unless added in very large amounts, acetone in water solution produces no effect on picrate solutions. The acetone blows off. If acetone is added after heating the picrate and carbonate mixture a color change occurs with smaller amounts.

The experiments summarized in Table 15 show the sensitiveness of the picrate solution to solutions of acetone added to the blood.

It will be observed that a distinct color change occurs in the picrate solution when a concentration of 2 c.c. of a solution of 0.05 mg. of acetone in 100 c.c. of water is added to 2 c.c. of normal blood. Placing the normal acetone content of the blood at 1 or 2 mg. per liter,[4] or

4. "Normal blood when analyzed as described for total acetone bodies yields only 1 or 2 mg. of precipitate, equivalent to from 0.013 to 0.026 gm. of acetone per liter. In diabetes as much as 2.5 gm. (250 mg.) of acetone bodies, calculated as acetone, has been observed, while patients under ordinarily good control show from 0.1 to 0.4 gm." (per liter).

TABLE 12.—Experiment 11. Dec. 8, 1919. The Effect of Pituitary Extract

2 c.c. water control	0.040
2 c.c. water + 1 c.c. surgical pituitary extract (pituitrin)*	0.115
2 c.c. water + 1 drop surgical pituitary extract (pituitrin)	0.040
2 c.c. water + a few crystals of chlorbutanol	0.342
2 c.c. water + powdered pituitary gland (P. D.)	0.040

* Surgical pituitary extract (pituitrin) is put up in chlorbutanol. Pituitary extract induced no change.

TABLE 13—Experiment 12. Thyroid Extract in Water

	Gm., Per Cent.
2 c.c. water control	0.040
2 c.c. water + 100 mg. powdered thyroid gland (P. D.)	0.100
2 c.c. water + 50 mg. powdered thyroid gland (P. D.)	0.080
2 c.c. water + 25 mg. powdered thyroid gland (P. D.)	0.060
2 c.c. water + 10 mg. powdered thyroid gland (P. D.)	0.040
2 c.c. water + 5 mg. pwodered thyroid gland (P. D.)	0.040

TABLE 14.—Experiment 12. Thyroid Extract in Blood

	Gm., Per Cent.
2 c.c. blood control	0.125
2 c.c. blood + 100 mg. powdered thyroid gland	0.200
2 c.c. blood + 50 mg. powdered thyroid gland	0.190
2 c.c. blood + 25 mg. powdered thyroid gland	0.165
2 c.c. blood + 10 mg. powdered thyroid gland	0.140
2 c.c. blood + 5 mg. powdered thyroid gland	0.130

Thyroid extract when added to blood induces a change.

TABLE 15.—Experiment 13. Acetone in Blood

Size of Blood Sample	Acetone		Blood Sugar Record, L.-B. Test			
	Amount Added	Strength of Solution	Series 1 May 1	Series 2 May 3	Series 3 May 4	Series 4 May 13
Control	.....	.....	0.113†	0.117‡	0.101§	0.103¶
2 c.c.	2 c.c.	*20 mg. to 100 c.c. water	0.250	0.210		
2 c.c.	2 c.c.	10 mg. to 100 c.c. water	0.220	0.195		
2 c.c.	2 c.c.	9 mg. to 100 c.c. water	0.220			
2 c.c.	2 c.c.	7 mg. to 100 c.c. water	0.198			
2 c.c.	2 c.c.	5 mg. to 100 c.c. water	0.185	0.180	.....	0.190
2 c.c.	2 c.c.	4 mg. to 100 c.c. wtaer	0.185	.....	.....	0.187
2 c.c.	2 c.c.	3 mg. to 100 c.c. water	0.180	.....	.....	0.193
2 c.c.	2 c.c.	2 mg. to 100 c.c. water	0.170	0.155	.....	0.189
2 c.c.	2 c.c.	1 mg. to 100 c.c. wtaer	0.150	.....	0.144	
2 c.c.	2 c.c.	0.75 mg. to 100 c.c. water	.....	0.140	0.143	0.172
2 c.c.	2 c.c.	0.50 mg. to 100 c.c. water	.....	0.135	0.135	0.170
2 c.c.	2 c.c.	0.25 mg. to 100 c.c. water	.....	0.125	0.128	
2 c.c.	2 c.c.	0.20 mg. to 100 c.c. water	.....	.....	0.127	0.144
2 c.c.	2 c.c.	0.10 mg. to 100 c.c. water	.....	.....	0.121	0.140
2 c.c.	2 c.c.	0.05 mg. to 100 c.c. water	.....	.....	0.114	

* Acetone C. P. dilutions based on specific gravity 0.797. Samples run with 100, 75, 50, 25, 15 and 10 mg. to 100 c.c. of water were practically black; no attempt was made to read them.
† Blood from a convalescent variola patient.
‡ Blood from a convalescent variola patient.
§ Blood from a normal person.
¶ The blood was taken from a boy who had an afebrile pyelitis.

from 0.001 to 0.002 mg. per cent., we are safe in saying that acetone interferes in the Lewis-Benedict test when the acetone content of the blood is increased 0.05 mg. per cent. above normal.

The remarkable sensitiveness of the picrate solution to acetone is better comprehended when we recognize the fact that in a 2 c.c. sample of blood to which is added 2 c.c. of a solution of 0.05 mg. of acetone to 100 c.c. of water, there is only 0.021 mg. (0.0000021 gm.) present.

TABLE 16.—SHOWING THE COMPARATIVE SENSITIVENESS OF THE LEWIS-BENEDICT TEST TO SOLUTIONS OF GLUCOSE IN BLOOD AND ACETONE IN BLOOD

Size of Blood Sample	Mg. Added	Record Blood + Glucose, Mg. Per Cent.	Record Blood + Acetone, Mg. Per Cent.		Record Water + Glucose, Mg. Per Cent.	Difference Between Blood + Glucose and Control, Mg. Per Cent.
2 c.c.	100	0.202	0.250		.....	0.094
2 c.c.	75	0.168	0.230		0.070	0.060
2 c.c.	50	0.152	0.220		**0.052**	0.044
2 c.c.	25	**0.114**	0.198		0.040	0.006
2 c.c.	10	0.108	0.185		0.040	0.000
2 c.c.	5	0.102	0.185		0.040	
2 c.c.	4	.....	0.180			
2 c.c.	3	.....	0.170			
2 c.c.	2	0.105	0.150			
2 c.c.	1	0.104	0.140	0.144		
2 c.c.	0.75	.....	0.135	0.143		
2 c.c.	0.50	.....	0.125	0.135		
2 c.c.	0.25	.....		0.128		
2 c.c.	0.20	.....		0.127		
2 c.c.	0.10	.....		0.121		
2 c.c.	0.05	.....		**0.114**	0.040	
Control	.....	0.108 May 15	0.113 May 13	0.101 May 4	May 15-20	

Heavy figures show limit of sensitiveness.

TABLE 17.—EXPERIMENT 14. MAY 15, 1920. GLUCOSE IN WATER

	Gm. Per Cent.
2 c.c. water control	0.040
2 c.c. water + 2 c.c. 100 mg. glucose - 100 c.c. water	Lost
2 c.c. water + 2 c.c. 75 mg. glucose - 100 c.c. water	0.070
2 c.c. water + 2 c.c. 50 mg. glucose - 100 c.c. water	0.053
2 c.c. water + 2 c.c. 25 mg. glucose - 100 c.c. water	0.040
2 c.c. water + 2 c.c. 10 mg. glucose - 100 c.c. water	0.040
2 c.c. water + 2 c.c. 5 mg. glucose - 100 c.c. water	0.040
2 c.c. water + 2 c.c. 2 mg. glucose - 100 c.c. water	0.040
2 c.c. water + 2 c.c. 1 mg. glucose - 100 c.c. water	0.040

It may be of interest to show the comparative sensitiveness of picrate solution to solutions of glucose and solutions of acetone. Table 16 is compiled from previous experiments.

The picrate solution is sensitive to glucose in a concentration of 25 mg. per cent., to acetone in a concentration of 0.05 mg. per cent. Glucose water solutions give distinct color change at 50 mg. per cent. (Table 17).

The picrate solution used in the Lewis-Benedict blood sugar method is 500 times more sensitive to acetone than it is to glucose.

TABLE 18.—Experiment 15. May 21, 1920. Diacetic Acid in Blood

Size of Blood Sample	Diacetic Acid		Blood Sugar Record, L.-B. Test
	Amount Added, Mg.	Strength of Solution	
2 c.c. control	...	.....	0.113
2 c.c.	100	Acetic-aceto-ester in 100 c.c. water	0.256
2 c.c.	80	Acetic-aceto-ester in 100 c.c. water	0.343
2 c.c.	60	Acetic-aceto-ester in 100 c.c. water	0.251
2 c.c.	50	Acetic-aceto-ester in 100 c.c. water	0.218
2 c.c.	40	Acetic-aceto-ester in 100 c.c. water	0.210
2 c.c.	30	Acetic-aceto-ester in 100 c.c. water	0.201
2 c.c.	20	Acetic-aceto-ester in 100 c.c. water	0.195
2 c.c.	10	Acetic-aceto-ester in 100 c.c. water	0.164
2 c.c.	5	Acetic-aceto-ester in 100 c.c. water	0.147
2 c.c.	5	Acetic-aceto-ester in 100 c.c. water	0.144
2 c.c.	0.75*	Acetic-aceto-ester in 100 c.c. water	0.128
2 c.c.	0.50	Acetic-aceto-ester in 100 c.c. water	0.127
2 c.c.	0.25	Acetic-aceto-ester in 100 c.c. water	0.115
2 c.c.	0.10	Acetic-aceto-ester in 100 c.c. water	0.115
2 c.c.	0.05	Acetic-aceto-ester in 100 c.c. water	0.113
2 c.c.	0.025	Acetic-aceto-ester in 100 c.c. water	0.112

* From this point on a different sample of blood was used. The record of the control reading was inadvertently omitted from the permanent record book. It was practically the same as the first control.

TABLE 19.—Experiment 15. Diacetic Acid in Water

Size of Blood Sample	Diacetic Acid		Blood Sugar Record, L.-B. Test
	Amount Added, Mg.	Strength of Solution	
2 c.c. control	...	.....	0.040
2 c.c.	100	Aceto-acetic-ester in 100 c.c. water	0.391
2 c.c.	80	Aceto-acetic-ester in 100 c.c. water	0.256
2 c.c.	60	Aceto-acetic-ester in 100 c.c. water	0.232
2 c.c.	50	Aceto-acetic-ester in 100 c.c. water	0.198
2 c.c.	40	Aceto-acetic-ester in 100 c.c. water	0.167
2 c.c.	30	Aceto-acetic-ester in 100 c.c. water	0.143
2 c.c.	20	Aceto-acetic-ester in 100 c.c. water	0.123
2 c.c.	10	Aceto-acetic-ester in 100 c.c. water	0.115
2 c.c.	5	Aceto-acetic-ester in 100 c.c. water	0.095
2 c.c.	4	Aceto-acetic-ester in 100 c.c. water	0.083
2 c.c.	2	Aceto-acetic-ester in 100 c.c. water	0.071
2 c.c.	1	Aceto-acetic-ester in 100 c.c. water	0.049

TABLE 20.—The Limit of Sensitivity of Picrate Solution

	Gm.		Gm.
To glucose added to water is	0.001	Added to blood	0.0005
To creatinin added to water is	0.00005	Added to blood	0.00012
To diacetic acid added to water is	0.00002	Added to blood	0.00001
To acetone added to water is	.....	Added to blood	0.000001
To epinephrin (adrenalin) added to water is	0.00004	Added to blood	0.0000005

TABLE 21.—The Picrate Sensitivity in Blood

To creatinin is approximately	4 times greater than to glucose
To diacetic acid is approximately	50 times greater than to glucose
To acetone is approximately	500 times greater than to glucose
To epinephrin (adrenalin) is approximately	1000 times greater than to glucose

In these experiments the aceto-acetic-ester is hydrolyzed to diacetic acid and ethyl alcohol in the process of the test. The diacetic acid is unstable and rapidly decomposes into acetone and carbon dioxid. Ethyl alcohol also influences the picrate solution. We are safe in making the statement that diacetic acid interferes when 0.25 mg. is added to 100 c.c. of blood or when 0.00002 gm. is added to a 2 c.c. sample of blood.

DISCUSSION

We have presented a series of experiments which tend to show that blood contains substances other than sugar which induce a color change in the picrate solution employed in the modified Lewis-Benedict blood sugar method. Under normal conditions, these substances may not interfere with the established normal range for this method. Under pathologic conditions several of these substances may interfere. Those which show the most marked influence are: epinephrin, acetone and diacetic acid. Creatinin may interfere, but does so in a less marked degree if we consider the comparative sensitiveness of the picrate solution to these substances.

As picrate solution reacts to smaller quantities of acetone than are normally present in the blood, the question may well be raised: do not the acetone bodies of the blood contribute to the established normal blood sugar range for the Lewis-Benedict test? Still another question may be asked: as epinephrin in infinitesimally small quantities induce a color change in the picrate solution, is it not possible that this substance when thrown into the general circulation, as is supposed to happen in emotional states, may induce a so-called hyperglycemia without mobilizing the glycogen stores of the liver?

PARALYSIS OF THE LEFT RECURRENT LARYNGEAL NERVE ASSOCIATED WITH MITRAL STENOSIS

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INTRODUCTION

A number of cases of paralysis of the left recurrent laryngeal nerve, associated with mitral disease, have been reported during the last twenty-three years. It seems probable, in view of the cases to be reported in this paper, that such a complication, as evidenced by aphonia or slight hoarseness, may be more common than is generally suspected. That this association is not more widely recognized may be due simply to the fact that the paralysis either is not noted or is not attributed to the underlying cardiac condition, and it is reasonable to suppose that, if the possibility of such a complication were borne in mind, more cases would come to light which might fit into this category.

The first mention of this condition in the literature is a report of two cases by Norbert Ortner [1] in 1897. We have found in all forty-four references on the subject, for which bibliographies can be found in the papers by Fetterolf and Norris,[2] Lian and Marcorelles,[3] Hall,[4] Permewan,[5] Lanza,[6] Carran,[7] and Quadrone.[8] In all sixty-one cases have been reported, the first thirty-seven of which have been collected and tabulated by Fetterolf and Norris in an excellent analysis of the subject. The largest single group of cases is that of eight, reported by Quadrone.

ETIOLOGY AND PATHOLOGY

The anatomic etiology of the paralysis has been the subject of some discussion, the view most widely held being that the enlarged left auricle causes pressure on the left recurrent nerve, producing either a neuritis or a pressure atrophy. In Ortner's two cases the nerve was atrophied. Fetterolf and Norris point out that the left auricular cavity expands readily only upward; posteriorly it expands against the aorta and the esophagus, anteriorly against the right auricle and the left ventricle, and below against the right auricle and the liver.

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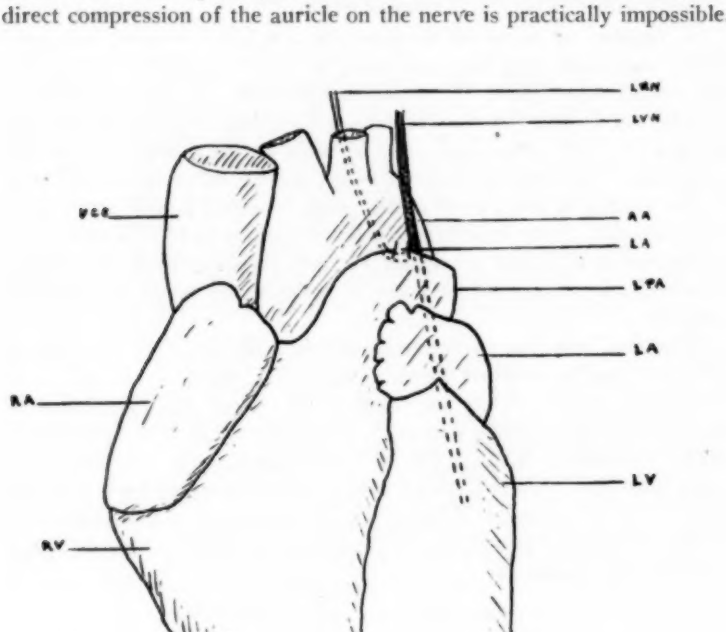
1. Ortner: Wien. klin. Wchnschr., No. 33, 1897.
2. Fetterolf and Norris: Am. J. M. Sc. **168**:625, 1911.
3. Lian, C., and Marcorelles, E.: Arch. d. mal. d. Coeur **6**:369, 1913.
4. Hall, de Haviland: Discussion at Roy. Med. Soc. London, Sect. Laryngol., May 2, 1913.
5. Permewan, W.: Ibid.
6. Lanza, L. R.: Gazz. d. osp., Milano **37**:65, 1916.
7. Carran, A., and Nunez, P. E.: Rev. med. d. Urugay, Montevideo **20**: 727, 1917.
8. Quadrone: Volumi di scritti Medici publ. in onore di Bozzolo, Torino, 1914.

Analysis of necropsy findings in the literature indicated to some authors that the etiology in seven cases was direct compression by the auricle or appendage; in two, persistent ductus arteriosus; in two, indirect compression acting on the pulmonary artery; and in one, cardiac displacement. Fetterolf and Norris discredit the greater number of these cases, holding that, as is shown in formalin-hardened subjects, direct compression of the auricle on the nerve is practically impossible.



VCS, superior vena cava; RA, right atrium; RV, right ventricle; LRN, left recurrent nerve; LVN, left vagus nerve; AA, aortic arch; LA, aortic ligament; LPA, left pulmonary artery; LA, left auricle; LV, left ventricle.

The accompanying diagram shows the relation of the structures implicated, and clearly bears out the point made by these authors. They conclude that the indirect mechanism may be variable, but when compression is accountable for paralysis, that it must always be caused by the nerve being squeezed between the left pulmonary artery and the aorta or aortic ligament, and that the paralysis is due to a pressure neuritis rather than actual destruction of the nerve.

Lian and Marcorelles set forth some of the various views on etiology which have been maintained. Ortner believed it was compression of the nerve against the aortic arch by the big left auricle; Kraus held it to be elongation of the nerve resulting from the right ventricular dilatation; Alexander, and later Fetterolf and Norris, claimed that dilatation of the left auricle acts by crowding back the pulmonary artery which is often dilated. The last named authors go on to state that in a few rare cases the voice troubles are the first symptoms which cause the patients to consult the physician who discovers the mitral stenosis. Of the eight cases of Quadrone, seven had eventually right as well as left recurrent laryngeal paralysis.

Lian and Marcorelles, Palasse, Boinet, Dmitrenko, Hofbauer, Alexander, Firschauer, and Osler used the right anterior oblique position at fifty degrees in fluoroscopy to reveal the big left auricle hidden by the spine. Lian and Marcorelles found that left lateral decubitus increased the troubles of the voice by increasing the contact of the left auricle and the pulmonary artery. A new theory which they expounded gives as a cause for the paralysis a thrombus in the left auricle or a certain degree of chronic mediastinitis, thus providing sufficient hardness for pressure.

We agree with Fetterolf and Norris that the pulmonary artery must be an intermediate agent in causing pressure, and with Lian and Marcorelles that a thrombus or a mediastinitis may occasionally be the responsible factor. Probably auricular fibrillation is an important additional factor besides the increase in size of the auricle in mitral stenosis because in auricular fibrillation the auricle remains ballooned out, with the ready production of thrombi.

Lian and Marcorelles further point out the difficulties in differential diagnosis, sufficient to render suspicious all cases not followed by necropsy. Aortic aneurysm, syphilitic mediastinitis, tracheobronchial adenopathy, esophageal cancer, and infectious or toxic recurrent neuritis must be ruled out, the roentgen ray being very important in excluding tracheobronchial adenopathy and aortic dilatation.

REPORT OF CASES

The following nine cases have been collected from the records of the past eight years of the Massachusetts General Hospital; it is probable that others, with a less marked laryngeal paralysis, have passed unnoticed:

CASE 1 (No. 185274).—H. A. L., 27, male, married. Monotype operator. Admitted Oct. 2, 1912.

Diagnosis.—Mitral stenosis. Dilated left auricle and left recurrent laryngeal nerve paralysis.

Family History.—Negative.

Past History.—Scarlet fever as a child. Fourteen years ago laid up for six weeks with pain in various joints. No redness or swelling of joints, and no fever. Patient was athletic up to two years ago.

Present Illness.—Six months ago caught cold and became hoarse. This hoarseness has persisted without remission until the present time. At the same time, he began to have some dyspnea on exertion. He stopped work seventeen weeks ago on account of dyspnea. Appetite has been lost. Slight cough for four days.

Physical Examination.—Mucous membranes cyanotic. Right pupil larger than the left.

Heart: Apex impulse felt in fifth space, 11 cm. from the midsternal line. Upper border at the second rib. Right border of dulness 4.5 cm. from the midsternal line. A blowing diastolic murmur is heard at the apex, working up into a loud presystolic roll ending in a very sharp first sound. No systolic murmur. There is a palpable thrill at the apex. The pulmonary second sound is accentuated and reduplicated. The pulses are equal, regular and synchronous.

Lungs: Numerous whistling and piping râles heard over both lungs.

Throat: Paralysis of left vocal cord. Larynx red. Left arytenoid swollen.

Wassermann: Negative.

Roentgen Ray: Marked dilatation of left auricle. Considerable peribronchial thickening with enlarged bronchial glands.

October 29: Discharged slightly relieved. Hoarseness continued.

Nov. 28, 1913: Admitted again with relapse and herpes zoster. Hoarseness present.

Discharged relieved in January, 1914. Not heard from further.

Case 2 (No. 186950).—E. F. W., 53, female. Admitted Jan. 6, 1913.

Diagnosis.—Mitral stenosis. Auricular fibrillation. Left recurrent laryngeal nerve paralysis.

Family History.—Negative, except that one sister died of tuberculosis. Four brothers and sisters died in infancy.

Past History.—Scarlet fever in childhood. Thirty years ago questionable rheumatism in ankles and knees.

Present Illness.—Six years ago, after sudden exertion, the patient had great dyspnea and cough, with frothy, white sputum. Since that time has had dyspnea on exertion. For the last two or three years this has increased. Four years ago first noticed edema of the legs, less lately than at first. Six months ago began gradually to get hoarse. The hoarseness soon reached its maximum and has not increased since. Frequent palpitation and irregularity of the heart beat. Often has retching spells.

Physical Examination.—Mucous membranes cyanotic. Heart's apex seen and felt, diffuse and heaving, in the fifth space. Left border of dulness 16 cm. from the midline, 8 cm. to the left of the nipple. Right border of dulness, 3.5 cm. to right of midline. Sounds irregular and of poor quality. Marked systolic thrill at apex. First sound sharp. Diastolic roll at apex. Loud systolic murmur heard all over precordia, transmitted into axilla. Pulmonic second sound greater than aortic and accentuated. Pulses equal, of poor volume, low tension, and irregular in force and rhythm. Not all beats reach the wrist.

Lungs: Moist crackling râles at bases.

Liver dulness fifth space to 12 cm. below the costal border.

January 8: Discharged against advice. Not heard from further.

Case 3 (No. 192727).—C. H., aged 48, female; watchmaker. Admitted six times between 1902 and 1916. December, 1913.

Diagnosis.—Mitral stenosis. Auricular fibrillation. "Decompensation." Hypertrophy and dilatation of the heart. Left recurrent laryngeal nerve paralysis.

Family History.—Negative.

Past History.—Chorea at 6; scarlet fever and diphtheria also in childhood.

Present Illness.—History of dyspnea, precordial pain and palpitation for past seven years.

Physical Examination.—Deeply cyanotic.

Heart: Apex impulse in sixth space, 8 cm. beyond midclavicular line. At apex there is a thrill. There are a diastolic roll and a soft blowing systolic murmur, heard loudest at the apex. Absolute irregularity of rhythm. Liver extends to 4 cm. below the costal border.

Wassermann: Negative.

Roentgen Ray: The heart shadow is very much enlarged. A bulging in region of left auricle. Diaphragm is low and limited on both sides. Apex is in seventh interspace, 10.8 cm. to left of median line. Right border is 6 cm. to right of median line. Enlarged heart with dilatation of left auricle.

Throat Consultation: Paralysis of the left recurrent laryngeal nerve.

June, 1915: Out-Patient Department. Electrocardiogram: Auricular fibrillation and right ventricular preponderance.

June, 1916: Edema and aphonia.

July, 1916: In ward again.

Died November, 1916.

CASE 4 (No. 199598).—M. E. G., aged 40, female, married, housewife. Admitted Jan. 5, 1915.

Diagnosis.—Right pyopneumothorax. Mitral stenosis. Auricular fibrillation. Left recurrent laryngeal nerve paralysis. Chronic passive congestion of the liver.

Family History.—Father, one brother and one sister died of tuberculosis.

Past History.—Inflammatory rheumatism at 17, lasting off and on for ten years. Laid up in bed three or four times for from five to six weeks with this. Chronic cough since 17 with yellowish sputum. For fifteen years has tired very easily and has had dyspnea on exertion.

Present Illness.—Nine months ago, edema of ankles; has been in bed since. During past month dyspnea has increased and cough has become more troublesome. For five days has been hoarse.

Physical Examination.—Slight cyanosis. Right pupil larger than the left.

Heart: Apex beat seen and felt in the sixth space. Supracardiac dulness 11.5 cm. in first space, 9 cm. in second space. Systolic thrill at apex. First sound sharp. Short rough systolic murmur. Diastolic roll. Soft systolic murmur at fourth left interspace. Pulmonic second sound greater than aortic and accentuated. Absolute arrhythmia.

Lungs: Right apex dull in front and dull tympany below. Moderately dull behind. Left chest, compensatory resonance. Breath sounds almost absent on right. Liver edge 5 cm. below costal border, tender. Slight edema of feet.

Wassermann: Negative.

Roentgen Ray: Hydropneumothorax on right with displacement of heart to left. Radial tracing and electrocardiogram show auricular fibrillation, the ventricular rate slowed apparently by digitalis.

Throat Consultation: Complete paralysis of left recurrent laryngeal nerve.

Chest tapped several times and pus withdrawn.

January 11: Died suddenly.

Necropsy (3422).—Pyopneumothorax, right; chronic endocarditis of the mitral valve, with stenosis; hypertrophy and dilatation of the heart (414 gm.); infiltration of auricular myocardium by inflammatory cells; chronic passive congestion; hydropericardium; ascites; anasarca; arteriosclerosis of the aorta and its great branches; moderate arteriosclerosis of the pulmonary artery and its branches; cholelithiasis; old infarcts of one kidney; chronic pelvic peritonitis; chronic appendicitis; chronic pleuritis.

CASE 5 (No. 202569).—G. D. G., aged 12, male; admitted April 10, 1915.

Diagnosis.—Rheumatic heart disease with mitral stenosis. Adherent pericarditis. Left recurrent laryngeal nerve paralysis.

Family History.—Negative.

Past History.—No tonsillitis, rheumatic fever (?), or chorea.

Present Illness.—Nine years ago, as a punishment, was kept in a cold bath for half a day. After this was ill for some time, and the "body swollen." For the next two years was sick most of the time. For the next four years he was much improved. Two years ago began to have cough and dyspnea. One year ago the symptoms became worse; he was in the Children's Hospital from February to July. Three weeks ago began to have severe cough with moderate expectoration, dyspnea, palpitation, edema, and hoarseness.

Physical Examination.—Large obstructive tonsils. Moist sibilant râles all over both lungs.

Heart: Left border of dulness 10.5 cm. from midsternal line. Right border 3 cm. from midsternal line. Apex impulse shifts 2 cm. out in fifth space in left lateral position. First sound at apex snapping; long presystolic roll. At left of sternum to and fro friction rub. Short systolic murmur at second left space. Shifting dulness in flanks. Liver edge 1 cm. below costal border. Marked edema of legs. Blood pressure: Systolic, 110; diastolic, 80. Wassermann: negative.

Roentgen Ray: Seven foot plate. Apex is in the sixth space, 9.4 cm. to left of median line. Right border is 5.6 to right of median line. Total transverse diameter 15 cm. Length of heart 16.3 cm. Diameter at base 12.6 cm. Enlargement both sides of heart. Diminished pulsation. Limited excursion (?). Adherent pericarditis.

Electrocardiogram showed very large "P" deflection indicating auricular hypertrophy, normal rhythm, and right ventricular preponderance.

Throat Consultation: Partial left recurrent laryngeal nerve paralysis.

Readmitted June 21, 1915.

June 27. Twenty-six ounces clear fluid, specific gravity 1.016, removed from abdomen.

Discharged relieved, July 3.

Died at home, Oct. 1, 1915.

CASE 6 (No. 204293).—K. F., aged 26, female, single. Admitted Sept. 18, 1915.

Diagnosis.—Mitral stenosis. Paralysis of left recurrent laryngeal nerve.

Family History.—Negative.

Past History.—Measles, whooping cough, chickenpox in infancy. Many sore throats up to tonsillectomy eleven years ago. Bronchitis and grippe one year ago.

Present Illness.—Sudden attack of dyspnea three years ago with palpitation and orthopnea. Coughed a great deal; yellow sputum with blood. Since then frequent dyspnea on exertion and often orthopnea. Occasional dull pain over heart on deep breathing. Legs ache and fingernails become blue on exertion. Over-exertion induces nausea and vomiting. Since bronchitis ten months ago has had a feeling of constriction in throat and hoarseness.

Physical Examination.—Slight cyanosis.

Heart: Apex impulse seen and felt in fifth space 10 cm. to left of median line. Left border of dulness 10 cm. to left, right border 3 cm. to right of median line. Upper border at the third rib. Supracardiac dulness 5 cm. wide. Sounds regular, of fair quality. Pulmonic second sound greater than the aortic and accentuated. First apical sound very loud. There is a long diastolic murmur with presystolic accentuation in the third left interspace and at apex. The first and second sounds over the whole precordia are reduplicated.

Slight edema of the shins.

Blood Pressure: Systolic, 100; diastolic, 72.

Wassermann: Negative.

Roentgen Ray: Seven foot plate and fluoroscopy. Enlargement of both sides of the heart. Heart pulsations indistinct. About the second interspace there is a bulging from the heart shadow which does not correspond to the aortic arch and is probably the left auricle. The left lower chest is slightly duller than the right. No definite process in lung. Enlargement of heart. Apex is 10.5 cm. to left of median line. Right border 3.5 cm. Total transverse diameter 14 cm. Greatest transverse diameter of great vessels 4.5 cm. Length of heart 16 cm. Diameter at base 13.5 cm.

Electrocardiogram shows right ventricular preponderance and auricular hypertrophy (mitral stenosis). Normal rhythm.

Throat Consultation: Left recurrent laryngeal nerve paralysis.

September 29: Discharged unrelieved to Out-Patient Department.

June 7, 1916: Patient writes that she is doing well.

August, 1917: Patient writes that she is sometimes short of breath, but as a whole is better. Not heard from further.

CASE 7 (No. 208447).—D. P., aged 12, male; admitted May 18, 1916.

Diagnosis.—Acute and chronic endocarditis of aortic and mitral valves. Left recurrent laryngeal nerve paralysis. Adhesive pericarditis (?).

First entry, July 28, 1915.

Family History.—Negative.

Past History.—Negative

Present Illness.—Four weeks before first entry into hospital became very feverish and had a bad headache. In bed three weeks with dyspnea, orthopnea and palpitation. Some pain, tenderness and swelling in left knee one week ago.

Physical Examination.—Apex impulse seen and felt in fifth space 11.5 cm. from midline. Presystolic thrill at apex and palpable shock attending pulmonic second sound. Broadbent's sign present. Marked cervical and suprasternal pulsation. Left border of dulness 11.5 cm. from midline. Diastolic, presystolic and systolic murmurs heard at apex. Harsh systolic and diastolic murmurs at second left interspace and over sternum. Corrigan pulses, capillary pulse, and pistol shot in groin. Blood pressure: systolic, 144; diastolic, 10 (?). Polygram shows a long a-c interval, indicating earliest grade of block.

Roentgen Ray: Seven foot plate. Apex is 10.4 cm. to left of median line. Right border 4.3 cm. to right of midline. Total transverse diameter of great vessels is 6 cm. Length of heart, 16.7 cm. Diameter at base, 12.3 cm. Enlargement of heart especially to left.

Wassermann: negative.

Electrocardiogram: Left ventricular preponderance. Normal rhythm.

Throat Consultation: Complete paralysis of left laryngeal nerve.

Symptoms increased and there was a severe epistaxis previous to second admission. Grew rapidly worse at second entry and was taken home to die. Not heard from further.

CASE 8 (No. 214292).—A. D., aged 15, female; admitted April 11, 1917.

Diagnosis.—Rheumatic heart disease with involvement of mitral and (?) tricuspid valves. Auricular fibrillation. Adherent pericarditis (?). Left recurrent laryngeal nerve paralysis.

Family History.—Negative.

Past History.—Measles at 11.

Present Illness.—One year and nine months ago had a cold, sore throat, swollen joints, and pains all over. In bed most of time for three months and symptoms cleared up, but has had dyspnea on exertion since that time. Four months ago became more dyspneic and had edema of feet. Dyspnea has been increasing. Frequent palpitation for two years and orthopnea for three months. Four months ago, both elbows swollen, tender and painful. Three days ago edema of legs and feet began, and for three days a brassy, unproductive cough.

Physical Examination.—Heart: Apex impulse seen and felt in sixth space, 10 cm. to left of midline, in mid-axillary line, 2.5 cm. outside nipple line. Systolic thrill and heaving impulse over precordium. Left border of dulness 10 cm. to left of midline; right border 6 cm. to right; supracardiac dulness, 7 cm. Upper border of dulness at second rib. First sound not heard at apex. Aortic second sound not heard; pulmonic second sound reduplicated and accentuated. Second sound at apex preceded by loud musical systolic murmur, and followed by a short diastolic murmur. At aortic area a short blowing systolic murmur was heard. At the pulmonic area a fine musical systolic murmur. To the right of the sternum in the fifth space is a musical systolic murmur transmitted to the right axilla. Pulses small, of low tension, absolutely irregular. Liver edge, 6 cm. below the costal border. Edema over the shins. Slight cyanosis.

Wassermann: negative.

Roentgen Ray: Seven foot plate. Apex 10 cm. to left of midline. Greatest transverse diameter of great vessels 6 cm. Right border 6.5 cm. from midline. Total transverse diameter 16.5 cm. Length of heart, 16.5 cm. Diameter at base 14 cm. General enlargement of heart shadow.

Electrocardiogram: Auricular fibrillation. Right ventricular preponderance.

Throat Consultation: Paralysis of left vocal cord.

Continued with "poor compensation" until May, 1918, when he was again admitted to the ward with "decompensation" and died the following day. No necropsy.

CASE 9 (No. 232912).—M. B., aged 22, female, married; admitted Sept. 29, 1919.

Diagnosis.—Mitral stenosis. Aortic regurgitation. Hypertrophy and dilatation of the heart. Paralysis of left recurrent laryngeal nerve.

Family History.—Negative

Past History.—"Double pneumonia" at birth. Many attacks of bronchitis. "Double pneumonia" at 14 with "weak heart" following. Gradual loss of weight for over a year.

Present Illness.—Felt well up to about a year ago when she became "run down." Eyesight became blurred and she had dizzy spells. Six months ago she caught cold, and raised thick yellow sputum which was occasionally blood-tinged. This lasted for six weeks. Her voice has been lost for about two months. Previously she had been hoarse for some time. She now has dyspnea on exertion and orthopnea.

Physical Examination.—Heart: Supracardiac dulness 6 cm. Left border of dulness 10 cm. to left of midline; midclavicular line 7 cm. from midline; right border of dulness 3.5 cm. to right of midsternum. Forceful impulse felt in fifth space 10 cm. to left of midline; felt also in suprasternal notch. Sounds regular, rapid and of good quality. Pulmonic second sound greater than the aortic second sound and accentuated. A long presystolic roll at the apex is followed by a sharp first sound, then a short systolic murmur. The second sound is heard. At the base are a short systolic murmur, and a short diastolic whiff, best heard over the third left interspace. The aortic second sound is heard. There is a presystolic thrill at the apex. Pulses equal, synchronous and of good volume and tension. No edema. Blood pressure: systolic, 96; diastolic, 53.

Wassermann: negative.

Roentgen Ray: Seven foot plate. The heart border is 5 cm. to the right of the median line and 7.8 cm. to the left. Total transverse diameter 12.8 cm. Length of heart 14.8 cm. Base, 11.8 cm. Total transverse diameter of great vessels is 4.4 cm. Both auricular regions are very prominent. The distance across the base is much increased. Width is somewhat increased. Appearance is that of mitral disease.

Electrocardiogram: Normal rhythm. Tachycardia. Right ventricular preponderance, and large P wave indicative of mitral stenosis.

Throat Consultation: Left vocal cord paralysis. No ulceration or other evidence of disease. October 11. Discharged unrelieved. Not heard from further.

DISCUSSION

Analysis of these nine cases shows that in Cases 1, 3 and 6 enlargement of the left auricle was shown by the roentgen ray. The right pupil was greater than the left in Cases 1 and 4. Electrocardiograms were taken in Cases 3, 4, 5, 6, 7, 8 and 9; in Cases 5, 6 and 9 auricular hypertrophy and right ventricular preponderance were found. Auricular fibrillation was present in Cases 2, 3, 4 and 8. Cases 3 and 8 also showed right ventricular preponderance by electrocardiogram. Case 4 had an extracardiac condition in the chest, displacing the entire heart. This was also the only case in which the mitral stenosis was proved by necropsy. Cases 5, 7 and 8 had possibly adherent pericarditis as well as mitral lesions. Case 2 was not roentgenographed.

The duration of the hoarseness or aphonia in the cases seen at the Massachusetts General Hospital varied from five days to ten months when the patients entered the wards; there was occasionally history of remissions.

SUMMARY

A series of nine cases of paralysis of the left recurrent laryngeal nerve associated with mitral stenosis has been found at the Massachusetts General Hospital during the last eight years. The record of but sixty-one cases in the literature, not including these nine cases, indicates that the condition is probably frequently overlooked.

THE BLOOD PICTURE BEFORE AND AFTER GOETSCH EPINEPHRIN TEST

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The basis of the findings recorded here are the observations made in thirteen cases in which the Goetsch epinephrin test was applied. These cases were suggestive of thyroid disease, diagnosed as such by Drs. Baetjer and Miller. The work was carried on simultaneously with that of Cowie, and the results obtained were similar, except that Cowie's work was not done in cases diagnosed as hyperthyroidism and, therefore, did not show such a lymphocyte increase.

One of the early blood changes in toxic goiter is an increase in the ratio of lymphocytes and polymorphonuclear leukocytes and also an actual rise in the percentage of lymphocytes.

It has been shown by other observers that epinephrin administered subcutaneously will increase the leukocyte count and the erythrocyte count. In this experiment, however, the erythrocyte increase was not dwelt on. The main object was to show the change which takes place in the differential leukocyte count.

Inasmuch as epinephrin augments many of the symptoms of toxic goiter—such as rapid pulse, increased blood pressure, marked tremor, extreme weakness, sensation of heat—and produces a shaky feeling over the entire body, symptoms similar to those produced by a strong stimulant, it was thought interesting to study the blood changes after the Goetsch test and see whether the blood picture characteristic of hyperthyroidism was augmented as were the other symptoms.

This was found to be the case. Slight toxic goiter cases, with nearly normal blood picture, presented the picture of an advanced or well marked case after the administration of epinephrin.

The results compared with those of Cowie's observations on various types of persons, five of whom were normal, showed a much more marked change.

Two interesting features were brought out in this experiment: 1. The rapidity with which the blood picture was altered (in less than one hour). 2. That in practically all borderline cases, the blood picture was of normal proportions or was preceded by a leukocytosis.

The results also showed that it is evident that the blood changes under normal conditions must be, in part at least, under control of the glands of internal secretion or of the autonomic nervous system or both.

REPORT OF CASES

CASE 1.—Before the administration of 8 minims of epinephrin, the white count was 3,400. The differential count was: polymorphonuclears, 61.2 per cent.; eosinophils, 2.4 per cent.; basophils, 1.2 per cent.; small lymphocytes, 24.8 per cent.; large lymphocytes and transitionals, 10.4 per cent.

One hour later the blood count was: leukocytes, 9,800. Differential count: polymorphonuclears, 44.8 per cent.; eosinophils, 1.2 per cent.; basophils, none; small lymphocytes, 45.6 per cent.; large lymphocytes and transitionals, 8.4 per cent.

Comment: Leukopenia existed in this case before the administration of the test. The differential count was normal except for a mild basophilia. One hour after the test was made there was an increase of 6,400 leukocytes, with an absolute lymphocytosis (increase 20.8 per cent.).

CASE 2.—Before the administration of epinephrin, the leukocyte count was 5,000. The differential count was: polymorphonuclears, 60 per cent.; eosinophils, 2.4 per cent.; basophils, none; small lymphocytes, 31.2 per cent.; large lymphocytes and transitionals, 6.4 per cent.

One-half hour later, the white count was 11,000. The differential count was: polymorphonuclears, 65.2 per cent.; eosinophils, 0.4 per cent.; basophils, none; small lymphocytes, 29.2 per cent.; large lymphocytes and transitionals, 5.2 per cent.

Comment: It is noted here that while there is an increase of 6,000 in the leukocyte count there is a reduction of 2 per cent. in the lymphocytes. This patient was markedly hyperthyroid and had a mild lymphocytosis from the beginning. She reacted quite markedly to the epinephrin test, and was quite exhausted at the end of one hour, so that it was thought best not to subject her longer to the inconveniences of taking pulse, blood pressure, etc. Therefore, the count was taken at the end of one-half hour instead of the usual hour test. It is evident that the blood film was taken while the cells were undergoing the change.

CASE 3.—Before the administration of epinephrin, the leukocyte count was 8,200 and the differential count was as follows: Polymorphonuclears, 61.6 per cent.; eosinophils, 0.8 per cent.; basophils, 0.8 per cent.; small lymphocytes, 25.2 per cent.; large lymphocytes and transitionals, 11.6 per cent.

Three-quarters of an hour later, the leukocyte count was 8,100 and the differential count was: polymorphonuclears, 51.6 per cent.; eosinophils, 0.8 per cent.; basophils, 0.8 per cent.; small lymphocytes, 33.6 per cent.; large lymphocytes and transitionals, 13.2 per cent.

Comment: In this case there is no increase in the leukocyte count. There was a subsequent rise of 8.4 per cent. in the lymphocytes.

CASE 4.—Before the administration of epinephrin, the leukocyte count was 9,400 and the differential count was as follows: Polymorphonuclears, 58.8 per cent.; eosinophils, 2 per cent.; basophils, 0.4 per cent.; small lymphocytes, 29.6 per cent.; large lymphocytes and transitionals, 9.2 per cent.

Three-quarters of an hour later, the leukocyte count was 13,000 and the differential count was: polymorphonuclears, 47.2 per cent.; eosinophils, 2 per cent.; basophils, 0.4 per cent.; small lymphocytes, 42.4 per cent.; large lymphocytes and transitionals, 8 per cent.

Comment: There was a rise of 3,600 in the leukocyte count and a very marked increase in the lymphocytes—13.6 per cent.

Case 5.—Before administration of epinephrin, the leukocyte count was 9,000 and the differential count was as follows: Polymorphonuclears, 70.8 per cent.; eosinophils, 0.8 per cent.; basophils, 0.4 per cent.; small lymphocytes, 20.4 per cent.; large lymphocytes and transitionals, 7.6 per cent.

One hour after the test was made, the leukocyte count was 13,000 and the differential count was: polymorphonuclears, 65.2 per cent.; eosinophils, 1.2 per cent.; basophils, 0.4 per cent.; small lymphocytes, 28 per cent.; large lymphocytes and transitionals, 5.2 per cent.

Comment: In this case there was an increase of 4,000 in the leukocyte count and a rise of 7.6 per cent. in the lymphocytes.

Case 6.—Before the administration of epinephrin, the leukocyte count was 9,000 and the differential count was as follows: Polymorphonuclears, 46 per cent.; eosinophils, 2 per cent.; basophils, 0.4 per cent.; small lymphocytes, 41.6 per cent.; large lymphocytes and transitionals, 10 per cent.

Three-fourths of an hour later, the leukocyte count was 14,800 and the diffdifferential count was: polymorphonuclears, 48 per cent.; eosinophils, 0.8 per cent.; basophils, none; small lymphocytes, 47.2; large lymphocytes and transitionals, 4 per cent.

Comment: This patient was definitely hyperthyroid. There was a rise of 5,800 in the leukocyte count and an increase of 5.6 per cent. in the lymphocytes.

Case 7.—Before the administration of epinephrin, the leukocyte count was 7,800 and the differential count was as follows: Polymorphonuclears, 75.2 per cent.; eosinophils, 0.4 per cent.; basophils, none; small lymphocytes, 17.2 per cent.; large lymphocytes and transitionals, 7.2 per cent.

Three-quarters of an hour later, the leukocyte count was 12,200 and the differential count was: polymorphonuclears, 68.8 per cent.; eosinophils, 0.4 per cent.; basophils, none; small lymphocytes, 26.8 per cent.; large lymphocytes and transitionals, 4 per cent.

Comment: In this case the leukocyte increase was 4,400 and the lymphocyte increase was 9.6 per cent.

Case 8.—Before the administration of epinephrin, the leukocyte count was 8,400 and the differential count was as follows: Polymorphonuclears, 56.4 per cent.; eosinophils, 4.8 per cent.; basophils, 0.8 per cent.; small lymphocytes, 28.8 per cent.; large lymphocytes and transitionals, 9.2 per cent.

One-half hour later, the leukocyte count increased to 11,800 and the differential count was: polymorphonuclears, 57 per cent.; eosinophils, 3.5 per cent.; basophils, 1.5 per cent.; small lymphocytes, 30 per cent.; large lymphocytes and transitionals, 8 per cent.

Comment: In this case there was a moderate constitutional reaction to the epinephrin. The increase in the leukyocyte count was 3,400 and 1.2 per cent. in the lymphocytes.

CASE 9.—Before the administration of epinephrin, the leukocyte count was 9,400 and the differential count was as follows: Polymorphonuclears, 68 per cent.; eosinophils, 1 per cent.; basophils, 1 per cent.; small lymphocytes, 20 per cent.; large lymphocytes and transitionals, 10 per cent.

Three-quarters of an hour later, the leukocyte count was 12,000 and the differential count was: polymorphonuclears, 65.2 per cent.; eosinophils, 1.2 per cent.; basophils, 0.4 per cent.; small lymphocytes, 27.2 per cent.; large lymphocytes and transitionals, 6 per cent.

Comment: There was a rise of 2,600 in the leukocyte count and an increase in the lymphocytes of 7.2 per cent.

CASE 10.—Before the administration of epinephrin, the leukocyte count was 6,800 and the differential count was as follows: Polymorphonuclears, 46.4 per cent.; eosinophils, 1.2 per cent.; basophils, 0.8 per cent.; small lymphocytes, 43.2 per cent.; large lymphocytes and transitionals, 8.4 per cent.

One-half hour later, the leukocyte count was 12,600 and the differential count was: polymorphonuclears, 39.6 per cent.; eosinophils, 1.2 per cent.; basophils, none; small lymphocytes, 54 per cent.; large lymphocytes and transitionals, 5.2 per cent.

Comment: There was an increase of 5,800 in the leukocyte count and a definite rise in the lymphocytes of 10.8 per cent.

CASE 11.—Before the administration of epinephrin the leukocyte count was 9,000 and the differential count was as follows: Polymorphonuclears, 64.4 per cent.; eosinophils, 0.4 per cent.; basophils, none; small lymphocytes, 29.2 per cent.; large lymphocytes and transitionals, 6 per cent.

One hour after the administration of epinephrin, the leukocyte count was 16,000 and the differential count was: polymorphonuclears, 46.4 per cent.; eosinophils, 1.6 per cent.; basophils, 0.4 per cent.; small lymphocytes, 42 per cent.; large lymphocytes and transitionals, 9.6 per cent.

Comment: There was a rise of 12.8 per cent. in the lymphocyte count, and an increase of 7,000 in the leukocyte count.

CASE 12.—Before the administration of epinephrin, the leukocyte count was 10,800 and the differential count was as follows: Polymorphonuclears, 68.8 per cent.; eosinophils, none; basophils, none; small lymphocytes, 23.2 per cent.; large lymphocytes and transitionals, 8 per cent.

Three-quarters of an hour after the administration of epinephrin, the leukocyte count was 16,400 and the differential count was: polymorphonuclears, 55.2 per cent.; eosinophils, 0.4 per cent.; basophils, 0.4 per cent.; small lymphocytes, 35.6 per cent.; large lymphocytes and transitionals, 8.4.

Comment: The increase in the leukocyte count was 5,600, and the rise in the lymphocytes equalled 12.4 per cent.

CASE 13.—Before the administration of epinephrin, the leukocyte count was 12,800 and the differential count was as follows: Polymorphonuclears, 64.8 per cent.; eosinophils, 0.8 per cent.; basophils, none; small lymphocytes, 26 per cent.; large lymphocytes and transitionals, 8.4 per cent.

One hour after the administration of epinephrin, the differential count was: polymorphonuclears, 48 per cent.; eosinophils, none; basophils, none; small lymphocytes, 47 per cent.; large lymphocytes and transitionals, 5 per cent. The leukocyte record was lost.

Comment: There was an increase of 21 per cent. in the lymphocyte count.

SUMMARY

The administration subcutaneously of from 4 to 8 minims of epinephrin will quickly produce a rise in the leukocyte count and cause an absolute lymphocytosis in hyperthyroid individuals. From this experiment, it is concluded that in toxic goiter an increase in the lymphocytes is not always present, especially in the borderline cases. A normal differential count is often found and in some cases a polymorphonuclear leukocytosis exists.

THE POSSIBLE PATHOGENICITY OF BACILLUS BOTULINUS *

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As commonly discussed in the literature, the danger of botulism lies entirely in the toxin produced by the organisms outside the animal body. Freed from toxin, *Bacillus botulinus* and its spores have frequently been fed or injected without loss of experimental animals. Warnings against spoiled food have commonly indicated that such foods were freed from all possible dangers of producing botulism by boiling.[1] The toxin is certainly destroyed by a brief heating at 80 C. or less. Certain strains of type B (for example the Nevin organism) and their spores are killed quickly at 100 C. More recently strains of type A (such as the Boise described by us in a recent paper [2]) have been shown to be very resistant. Clearly, a scrutiny of the organism itself as a possible source of danger was demanded. In examining suspected canned foods, *B. botulinus* has been found by us commonly associated with other resistant spore forming anaerobes, predominantly belonging to the series including *B. sporogenes* and *B. putrificus*. This group of "end-spore" formers are obligate anaerobes, produces putrefactive odors, and display so many reactions in common that animal experimentation has been necessary for the separation of the toxin forming *B. botulinus* from the less dangerous forms.

The association of these forms in food spoilage, and their common reactions establish the presumption of a common source or habitat. For some of them fecal or sewage pollution is clearly indicated. One of these (*B. sporogenes* [?]) isolated in this laboratory from well water and from food canned with it, was clearly traced to sewage of human origin. *B. botulinus* itself is reported by Kemper and Pollock,[3] and by Burke and Dickson,[4] as found in feces of swine not showing evidences of disease. Graham [5] traced cases of forage poisoning

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* From the Microbiological Laboratory, Bureau of Chemistry, in cooperation with the Pathological Division, Bureau of Animal Industry, U. S. Department of Agriculture, Washington, D. C. Published by permission of the Secretary of Agriculture.

1. Dickson: J. A. M. A. **69**:966 (Sept. 22) 1917.

2. Thom, Charles, Edmondson, Ruth B., and Giltner, L. T.: Botulism from Canned Asparagus, J. A. M. A. **73**:907 (Sept. 20) 1919.

3. Kemper and Pollock: Deutsch. med. Wchnschr. 505, 1897.

4. Burke, G. S.: The Occurrence of Bacillus botulinus in Nature, J. Bacteriol. **4**:54 (Sept.) 1919.

5. Graham, R., Himmelberger, L. R., and Pontius, R. L.: A Disease Resembling "Forage Poisoning" in Horses and Mules Wherein Oat Hay Incorporated the Primary Factor. Proc. U. S. Live Stock Sanit. Ass'n, 1915, pp. 22.

of horses to organisms belonging to type B of the *B. botulinus* group present in hen dung eaten with oats. The same results have been obtained here from hens experimentally fed with cultures of the Nevin organism by Buckley and Shippen.[6] The presence of occasional organisms of the *B. botulinus* group in the alimentary canal is thus clearly shown.

Shippen[7] working with the Nevin strain of *B. botulinus,* which he found to produce toxin freely at body temperature, records the conviction that under some conditions this strain might be expected to become pathogenic. Most of our previous experiments with feeding and inoculation of pure cultures of *B. botulinus* and its spores freed from toxin failed to produce the characteristic disease. A new series of experiments was suggested by the work of Bullock and Cramer,[8] who experimented with *B. welchii, B. tetani* and forms of similar morphology which fail to produce lesions when saline suspensions of pure cultures freed from toxin are used. They showed that if animals were inoculated with pure cultures of these forms and were injected also with small amounts of calcium chlorid, calcium nitrate, calcium acetate or sterile watery extracts of earth, lesions would develop and the characteristic toxic symptoms would follow. These authors attributed the result to the breaking down of the resistance of the host animal by the reagent injected. This phenomenon was designated "Kataplylaxis."

Our series of experiments after their method are given in detail as follows: In the first series two guinea-pigs (Nos. 1 and 2) were injected subcutaneously at the same site successively with heated spores of the Boise strain and calcium chlorid. The interval between injections was but a few minutes. The first animal received 10 mg. of calcium chlorid solution and 1 c.c. of a four months old culture of *B. botulinus* in plain milk, which had been heated at 80 C. for fifteen minutes to kill the vegetative forms and to destroy the soluble toxin. The second animal received 20 mg. of calcium chlorid and 1 c.c. of the heated spores. The check pig (No. 3) received 1 c.c. of the heated spores only. The first two guinea-pigs died of typical botulism, the first within five days, the second within four days. The check animal remained well. About twenty-four hours after the injections of calcium chlorid and spores, there was a slight swelling at the site of inoculation on all three animals. From the small ulcerlike local lesion

6. Buckley, J. S., and Shippen, L. P.: Preliminary Report on the Relation of Anaerobic Organisms to Forage Poisoning, J. Am. Veter. M. A. **50**:809, 1917.

7. Shippen, L. P.: Toxin Formation by a Variety of *B. botulinus* When Cultivated Aerobically under Various Conditions. Its Possible Production in the Animal Body, Arch. Int. Med. **23**:346 (March) 1919.

8. Bullock, W. E., and Cramer, W.: On a New Factor in the Mechanism of Bacterial Infection, Proc. Roy. Soc. Lond. Series B. **90**:513 (May 15) 1919.

of the animals which received calcium chlorid, virulent bacilli were recovered. Cultures from the liver and heart's blood of these guinea-pigs were negative for *B. botulinus*.

In the second series guinea-pigs, Nos. 4, 5 and 6, respectively, were injected at the same site at the same time with 20 mg. of calcium chlorid and 0.2 c.c. of heated *B. botulinus* spores from a dextrose beef infusion culture. The check animal received 0.2 c.c. of heated spores only. All three of the animals died of typical botulism, Nos. 4 and 6 within three days and No. 5 within four days.

In the third series of animals, Nos. 7, 8 and 9, the injections were made simultaneously at different sites, the sites were only from 3 to 4 cm. apart. The same doses were given these guinea-pigs as were given those in the above experiment. Guinea-pig No. 7 died within four days, and guinea-pig No. 8 within six days, but the check guinea-pig, No. 9, lingered on for two weeks. The organism was recovered in virulent form from the local lesion of guinea-pig No. 8, which died six days after the injection.

Spores freed from their toxin by washing were used in the fourth series. A culture of *B. botulinus* in dextrose beef infusion showing free spores was centrifuged to throw down the spores which were then washed fourteen times (in previous experiments we demonstrated the necessity of thorough washing for *B. botulinus* cells to free them completely from their toxin) in physiologic sodium chlorid solution. Two pigs (Nos. 10 and 11) were each injected successively at the same site with 0.2 c.c. of the saline spore suspension and 20 mg. of calcium chlorid. Both animals died at the end of three days. The check animal (No. 12) showed no symptoms until about the fifth day. It died on the fifteenth day of typical botulism. The local lesion and the liver of the two animals receiving calcium chlorid and spores were cultured, and virulent *B. botulinus* organisms were recovered from the local lesion only. The Boise strain was used in all of the above experiments.

In the fifth series spores from old liver broth cultures of the Nevin strain were washed twice in physiologic sodium chlorid solution, and the resulting spore suspension was heated at 80 C. for fifteen minutes, ahd a direct microscopic count was made by means of a Breed smear. There were 31,000,000 spores per c.c. One guinea-pig (No. 13) was then injected subcutaneously with 1 c.c. of the washed and heated spores mixed with 1 c.c. of a 2 per cent. calcium chlorid solution (20 mg.). Another animal (No. 14) was injected at different sites with the same quantities of spores and calcium chlorid. The sites were only a few centimeters apart. Another guinea-pig (No. 15) was injected with similar doses at the same site, but with an interval

of one hour between the injections of spores and calcium chlorid. Another pig (No. 16) was injected at the same site with similar doses of spores and calcium chlorid but with an interval of two and one-half days between the injections. In each case the spores were injected first. The check animal (No. 17) received an injection of 1 c.c. of the spores alone. The only animal in this series which showed typical symptoms of botulism and died was guinea-pig No. 16. Death occurred seven days after the first injection. The other animals were slightly "off" condition about a week after the injection, but this was probably the effect of the calcium chlorid. At necropsy, cultures were made from the liver and local lesion of guinea-pig No. 16. Virulent bacilli were recovered from the local lesion only.

The Boise strain was then used in Series 6. Old, liver broth cultures showing free spores were centrifuged, and the resulting sediment first washed twice in physiologic sodium chlorid solution and then heated at 80 C. for fifteen minutes. Direct microscopic count indicated 30,000,000 spores per c.c. One c.c. of the spore suspension mixed with 1 c.c. of a 2 per cent. calcium chlorid solution was then injected into a guinea-pig (No. 18). Guinea pig No. 19 was injected simultaneously with the same quantity of spores and calcium chlorid at different sites; guinea-pig No. 20 was injected at same site with an interval of one hour between the injections; and guinea-pig No. 21 at the same site with an interval of two days between the injections of spores and calcium chlorid. The check animal received 1 c.c. of spores only. Animals Nos. 18 and 20 respectively, began showing symptoms early, No. 20 within forty hours and No. 18 within forty-eight hours. The latter died four days later, but guinea-pig No. 20 died about forty-six hours after the injection. Guinea-pig No. 19 showed typical symptoms in three days, gradually got worse and died on the tenth day. Guinea-pig No. 21 and the check guinea-pig (No. 22) began showing symptoms on the third day. Six days after the injection guinea-pig No. 21 was very ill and died on the thirteenth day. The check pig died at about the same time also. At necropsy, cultures were made from the local lesion and liver of each animal. In each case virulent bacilli were recovered from the local lesions only.

The local lesion is apparently the effect of the calcium chlorid. When injected alone it produces a local inflammatory edema with more or less necrosis of the tissues at the point of injection. In some of the experimental animals there is an ulcer at the seat of injection which heals over in the course of a few weeks. The local lesion in the animals injected with both calcium chlorid and *B. botulinus* spores differs in no apparent way from that produced by the calcium chlorid

alone. There is practically no lesion where spores alone are injected. Smears made from the exudate of the local lesion of animals injected with spores and calcium chlorid showed generally only a few bacilli and many leukocytes. The leukocytes contained no bacilli. The finding of bacilli where spores were injected indicates that germination took place in the body. Smears from the local lesion of the check animal which died showed practically no organisms, but cultures revealed the presence of the bacilli in virulent form.

These experiments, which are summarized in Table 1, show that typical botulism was induced in every case where heated spores of the Boise strain were injected subcutaneously with small amounts (from 0.5 to 1.0 c.c) of a 2 per cent. calcium chlorid solution. All but two of the check pigs, which received spores only, died of botulism also, but the time interval between the deaths of those receiving spores and calcium chlorid and those receiving spores only was shortened considerably, apparently in the presence of the calcium salt. The Nevin strain gave negative results, on the whole, as only one animal died out of the four which were injected with spores and calcium chlorid. The check animal also lived. In most cases when the guinea-pigs were injected with spores and calcium chlorid at the same time at the same or different sites, death occurred in from three to six days. When two days elapsed between the injection of the spores and calcium chlorid, death was usually delayed several days.

The fact that all but two of the check animals, inoculated with spores only in the foregoing experiments died, suggested occasional pathogenicity unsupported by calcium chlorid. With this in view we repeated a number of our former experiments on the effect of the detoxified spores of *B. botulinus* in the animal body, enlarging the dose, and paying attention to the actual number of spores fed. Feeding was substituted for injection because botulism is primarily possible only by the consumption of the organisms or their toxin. These experiments given in detail are as follows.

SUMMARY OF EXPERIMENTS

Series 7.—An old culture of *B. botulinus* (Boise strain) in dextrose beef infusion showing free spores was centrifuged to throw down the spores, which were then washed three successive times in physiologic sodium chlorid solution. This saline spore suspension was heated at 80 C. for fifteen minutes, and 3 c.c. fed to a guinea-pig (No. 23). Feeding in all experiments was accomplished by introducing a pipet containing the spore suspension directly into the mouth of the animal. This pig showed no symptoms and remained well.

TABLE 1.—Effect of Injections of Calcium Chlorid and Toxin-free Spores of *B. botulinus*

Series of Experiments	Guinea-Pig No.	Subcutaneous Injections of		Strain	Site of Inoculation	Interval of Time between Injections	Death from Botulism
		$CaCl_2$ in Mg.	Spores				
1	1	10	1.0 (Heated at 80 C. for 15 Min.)	Boise	Same	1-2 Min.	5 days
	2	20	1.0 (Heated at 80 C. for 15 Min.)	Boise	Same	1-2 Min.	4 days
	3 (check)	0	1.0 (Heated at 80 C. for 15 Min.)	Boise	—	1-2 Min.	Remained well
2	4	20	0.2 (Heated at 80 C. for 15 Min.)	Boise	Same	0	3 days
	5	20	0.2 (Heated at 80 C. for 15 Min.)	Boise	Same	0	4 days
	6 (check)	0	0.2 (Heated at 80 C. for 15 Min.)	Boise	—	—	3 days
3	7	20	0.2 (Heated at 80 C. for 15 Min.)	Boise	Different	1-2 Min.	4 days
	8	20	0.2 (Heated at 80 C. for 15 Min.)	Boise	Different	1-2 Min.	6 days
	9 (check)	0	0.2 (Heated at 80 C. for 15 Min.)	Boise	—	—	14 days
4	10	20	0.2 (washed)	Boise	Same	1-2 Min.	3 days
	11	20	0.2 (washed)	Boise	Same	1-2 Min.	3 days
	12 (check)	0	0.2 (washed)	Boise	—	—	15 days
5	13	20	1.0 (washed and heated 31,000,000 to the c.c.)	Nevin	Same	0	Slight symptoms, recovered
	14	20	1.0 (washed and heated 31,000,000 to the c.c.)	Nevin	Different	1-2 Min.	Slight symptoms, recovered
	15	20	1.0 (washed and heated 31,000,000 to the c.c.)	Nevin	Same	1 hour	Slight symptoms, recovered
	16	20	1.0 (washed and heated 31,000,000 to the c.c.)	Nevin	Same	2½ days	7 days
	17 (check)	0	1.0 (washed and heated 31,000,000 to the c.c.)	Nevin	—	—	Remained well
6	18	20	1.0 (washed and heated 30,000,000 to the c.c.)	Boise	Same	0	4 days
	19	20	1.0 (washed and heated 30,000,000 to the c.c.)	Boise	Different	1-2 Min.	10 days
	20	20	1.0 (washed and heated 30,000,000 to the c.c.)	Boise	Same	1 hour	46 hours
	21	20	1.0 (washed and heated 30,000,000 to the c.c.)	Boise	Same	2 days	13 days
	22 (check)	0	1.0 (washed and heated 30,000,000 to the c.c.)	Boise	..	..	13 days

Another guinea-pig (No. 24) was fed with 6 c.c. of the heated spores. Six days later this guinea-pig was showing typical symptoms of botulism. It died eight days after the feeding.

Series 8.—Both the Boise and Nevin strains were used. Old cultures in liver bouillon were centrifuged to throw down the spores and the resulting sediment diluted with physiologic sodium chlorid solution and heated at 80 C. for fifteen minutes. A direct microscopic count of the saline suspension was then made by means of the Breed smear,

and 78,000,000 Nevin spores (1 c.c.) fed to one guinea-pig (No. 25) and 53,000,000 (1 c.c.) Boise spores fed to another guinea-pig (No. 26). The pig which received the spores of the Boise strain remained well, but the one which received the Nevin strain showed typical symptoms in five days and died within seven days.

Series 9.—The guinea-pigs (Nos. 27 to 32 inclusive) were fed with quantities of washed and heated spores of the two strains. The count for each strain was 50,000,000 spores per c.c. As checks two guinea-pigs for each strain were injected respectively with 1 c.c. of supernatant liquid from heated centrifuged spores and with 1 c.c. of unheated spores. Cultures made from the heated supernatant liquid showed slight growth after several days, showing that all the spores had not been thrown down in the centrifuge. However, for practical purposes, it was assumed that the liquid was free from spores. The Nevin pig (No. 27) which was fed 50,000,000 spores remained well but the Boise guinea-pig (No. 30) showed symptoms on the second day and died seventeen days after the feeding. Virulent bacilli were recovered from the liver of this animal. The check guinea-pigs which received supernatant liquid from heated, centrifuged spores remained well. The two checks on the virulence of the cultures, that is, the animals which received 1 c.c. of unheated spore suspension, died in about eighteen hours. The difference in time is striking.

Series 10.—Doses of 75,000,000, 100,000,000 and 112,500,000, respectively, of washed and heated spores from Nevin strain cultures were fed to guinea-pigs Nos. 33, 34, and 35, respectively, with no ill results. The check guinea-pig (No. 36) which received an intraperitoneal injection of 1 c.c. of supernatant liquid from the centrifuged heated spores also remained well.

Series 11.—The dose of spores was increased. Six c.c. of a washed and heated spore suspension from a culture of the Boise strain, containing 180,000,000 spores to the c.c. were fed to guinea-pig No. 37. The check, No. 38, received an intraperitoneal injection of 1 c.c. of the supernatant liquid from a portion of the centrifuged heated spores. Guinea-pig No. 37 died of typical botulism within five days but the check animal remained well. At necropsy a culture was made from the liver of guinea-pig No. 37 but the organism was not recovered.

Series 12.—In the next experiment small and large doses of Boise spores were fed to guinea-pigs Nos. 39 and 40 respectively. The small dose consisted of 32,000,000 spores in salt solution, the large dose consisted of 192,000,000. The check animal (No. 41) as before received 1 c.c. of supernatant liquid from a portion of the centrifuged, washed and heated spores. The guinea-pig which received the small

dose showed no symptoms but the one which received the large dose showed symptoms in about two days and died seven days after feeding. The check animal remained well.

TABLE 2.—Effect of Feeding Toxin-free Spores of *B. botulinus*

Series	Guinea-Pig	Strain of B. Botulinus	Dose of Heated Spores		Death from Botulism
			No. C.c.	No. Spores to C.c.	
7	23	Boise	8.0	No count was made	Remained well
	24	Boise	6.0	No count was made	8 days
8	25	Nevin	1.0	78,000,000	7 days
	26	Boise	1.0	53,000,000	Remained well
9	27	Nevin	1.0	50,000,000	Remained well
	28 (check)	Nevin	1.0	Injection of supernatant liquid from centrifuged spores	Remained well
	29 (check)	Nevin	1.0	Injection of 50,000,000 unheated spores	18 hours
	30	Boise	1.0	50,000,000	17 days
	31 (check)	Boise	1.0	Injection of supernatant liquid from centrifuged spores	Remained well
	32 (check)	Boise	1.0	Injection of 50,000,000 unheated spores	18 hours
10	33	Nevin	1.5	75,000,000	Remained well
	34	Nevin	2.0	100,000,000	Remained well
	35	Nevin	2.25	112,500,000	Remained well
	36 (check)	Nevin	1.0	Injection of supernatant liquid from centrifuged spores	Remained well
11	37	Boise	6.0	180,000,000	5 days
	38 (check)	Boise	1.0	Injection of supernatant liquid from centrifuged spores	Remained well
12	39	Boise	0.5	32,000,000	Remained well
	40	Boise	3.0	192,000,000	7 days
	41 (check)	Boise	1.0	Injection of supernatant liquid from centrifuged spores	Remained well

In general, the experiments with the Boise strain were consistent. Small doses of heated spores produced no effect when fed to guinea-pigs but large doses promptly caused botulism. Again the experiments with the Nevin organism were unsatisfactory. In one case a dose of only 78,000,000 spores caused death but in other cases doses of 100,000,000 and 112,500,000 spores failed to produce botulism. These results, summarized in Table 2, while irregular, confirm the discrepancies in our former experiments with the detoxified bacilli or spores of *B. botulinus*. They show that occasionally the guinea-pigs will develop the specific disease and die when fed or injected with large doses of the spores freed from toxin in routine manner. Just what conditions in the animal body favor the growth of the organism are not known. It is quite possible that botulism, when it occurs in such cases, is the result of the absorption of intracellular toxin which is protected during the heating process by the highly insulated spore wall and released in the body upon the dissolution of the spore wall, or,

on the other hand, it may be that the spores actually germinate in the animal body, probably in the small intestine and produce the toxin there. Experiments on this phase of the question are contemplated.

GENERAL DISCUSSION

The fundamental importance of knowing more about these toxin-forming anaerobes is brought out by these results. The line between pathogenicity and saprophytism is not a sharp one. Our conception of species in bacteria is equally hazy. *Bacillus botulinus* as a species very clearly consists of a series of strains or varieties with a common morphology and a common ability to produce particular toxins. These strains differ in details of reactions. Van Ermengem evidently possessed a form which produced toxin slightly, if at all, at body temperature. The Nevin organism (type B) as studied by Shippen grows and produces toxin freely at 37 C. Both are equally easily killed by boiling. The Boise strain, one of the Type A series, is resistant to high temperatures and in addition finds its optimum development at body temperature.

This combination of toxin forming power and optimum growth at body temperature called for continued investigation of the possibilities of pathogenicity for some at least of these strains. Although the organism was considered a strict saprophyte, occasional deaths of check animals occurred under conditions difficult to interpret. Search for explanation of these cases led to the experiments here reported. It has been shown that *B. botulinus* will produce botulism in experimental animals if calcium chlorid is injected with toxin-freed spores as described by Bullock and Cramer, for *B. welchii* and its allies. In this respect, therefore, *B. botulinus* reacts in the same way as the organisms of gas gangrene and tetanus studied by these authors. A limited pathogenicity is thus established.

This increased evidence of pathogenicity led to the subsequent series of feeding experiments, using large quantities of *B. botulinus* spores. The large number of deaths among these experimental animals gives additional warning of the dangerous character of the organism itself.[9] These results decrease our confidence in the effect of ordinary heating as a means of freeing suspected foodstuffs from possible danger.

SUMMARY AND CONCLUSIONS

1. Calcium chlorid acts as an accessory factor in producing botulism when injected into guinea pigs with toxin-freed bacilli or spores of

9. These results obtained independently are in agreement with those of Paul F. Orr: Some Observations on the Biological Characteristics of *B. botulinus* and Its Toxins, Abstr. Bacteriol. **4**: (Feb.) 1920.

B. botulinus. Under such conditions the behavior of *B. botulinus* is similar to that reported by Bullock and Cramer for the organisms of gas gangrene and tetanus.

2. On the basis of experiments tried, toxin-freed spores of the Boise strain of *B. botulinus*, when fed in sufficiently large doses to guinea-pigs, can produce the specific disease and death, while the Nevin strain similarly fed does so occasionally. A limited pathogenicity is thus indicated.

3. Foods suspected of containing *B. botulinus* should be destroyed, not heated and eaten.

A STUDY OF THE RENAL CONCENTRATION POWER FOR URIC ACID IN EARLY CHRONIC INTERSTITIAL NEPHRITIS

SECOND PAPER

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In a previous communication we presented a preliminary report of our work regarding the concentration power of the kidney for uric acid in early chronic interstitial nephritis. By the term interstitial nephritis was meant the renal anatomic lesion which is commonly associated with that condition. The cases presented as positive nephritis were only such as could readily be diagnosed by the conventional methods. It was shown that in such the renal concentration power for uric acid was markedly lowered, while in normal individuals no such lowering took place. A number of cases were also recorded in which a positive diagnosis of nephritis could not be made by the conventional methods. These cases, however, gave many suspicious symptoms of nephritis, and were, therefore, listed as questionable nephritis. They all showed lowering of the renal function for uric acid, and the thought was expressed by us that they were in reality positive nephritis as their further development would prove. In continuing the general study, we have made observations of the renal concentration power for uric acid in a number of diversified conditions. It has developed that the findings thus obtained are of crucial importance in the application of the method to the diagnosis of early chronic interstitial nephritis. A consideration of their significance is hence now undertaken.

METHOD OF COMPUTING RENAL CONCENTRATION POWER

In our preliminary work the renal concentration power was computed by merely dividing the amount of urine uric acid per hundred c.c. by the amount of blood uric acid per hundred c.c., both specimens having been taken simultaneously. The renal concentration power for any given substance is represented by the concentration of that substance in the urine. The concentration of any substance in a fluid is represented by the amount of that substance which is contained in a given volume of the fluid. In our original method of computation, the volume of urine was kept stationary at 100 c.c., the amount of

uric acid in such a volume varying as the conditions altered. It is evident that such a method of computation might be liable to certain inaccuracy since only one of the factors concerned in concentration is variable (namely, the amount of uric acid), the other factor (namely, the volume of fluid) being kept stationary. In order to obviate this error we have in the present communication adopted the formula recently proposed by Van Slyke for the above purpose. We do not here enter into a discussion of the Van Slyke method of computation but refer all interested to his original communication.[1]

In comparing Van Slyke's formula with our original method in the cases herein reported, a material difference in result occurred in about 10 per cent. of the cases.

AMOUNT OF BLOOD URIC ACID NOT A GUIDE TO THE RENAL FUNCTIONAL ACTIVITY FOR URIC ACID

In our preliminary communication we drew attention to the fact, which was deducted from the cases therein reported, that the mere amount of blood uric acid was not necessarily a guide to the renal functional activity for uric acid. This, of course, means that estimations of blood uric acid are unreliable for the judgment of the uric acid renal function. We desire here to emphasize this point and to add further proof thereof.

Table 1 shows a number of cases in which the blood uric acid was high, but the blood renal uric acid co-efficient was normal, showing a normal renal function for uric acid. This table further shows that when these cases were placed on a purin free diet, the blood uric acid promptly fell to normal.

TABLE 1.—CASES WITH NORMAL RENAL FUNCTION FOR URIC ACID

Patient	Ordinary Diet		Purin Free Diet	
	Blood Uric Acid Mg. per 100 c.c.*	Coefficient†	Blood Uric Acid Mg. per 100 c.c.*	Coefficient†
H. W. H.	6.47	2.6	2.90	2.8
C. M.	6.82	2.9	2.87	3.1
F. G.	7.20	3.2	3.00	3.7
C. A.	7.67	3.8	2.60	4.6
A. A.	5.80	2.6	2.10	2.9

* Normal blood uric acid per 100 c.c., 1 to 3 mg.
† Normal coefficient, 2.4 to 4.7 mg.

Table 2 contains cases in which the blood uric acid was high, but in which the blood renal uric acid co-efficient was low, showing a reduced

1. Proceedings of the American Society of Experimental Medicine and Biology, December, 1919.

renal function for uric acid. Table 2 also shows that when these cases were placed on a purin free diet, the blood uric acid was not brought to normal.

TABLE 2.—CASES WITH REDUCED RENAL FUNCTION FOR URIC ACID

Patient	Ordinary Diet		Purin Free Diet	
	Blood Uric Acid Mg. per 100 c.c.*	Coefficient†	Blood Uric Acid Mg. per 100 c.c.*	Coefficient†
F. W.	6.62	1.4	4.97	1.5
F. W.	7.94	0.92	6.01	0.96
A. D.	7.32	0.94	6.30	0.87
M. L.	6.46	1.1	5.20	1.3
W. H.	6.96	1.6	5.15	1.7

* Normal blood uric acid per 100 c.c., 1 to 3 mg.
† Normal coefficient, 2.4 to 4.7 mg.

The response and nonresponse of the blood uric acid to diet in these two classes of cases appears to us to be absolute proof that the blood renal uric acid co-efficient is the correct index of the renal functional activity for uric acid and that the mere blood amount of uric acid is no such guide. Both classes gave high figures for blood uric acid and from these figures alone it could not have been judged whether or not there was present a reduced renal uric acid function. We feel that this point needs emphasis because it has been and is now the prevalent practice on the basis of mere blood estimations of uric acid to make diagnosis for or against a reduction of renal function. It is certain such procedure frequently leads to erroneous conclusions which have confused the clinician and caused dissatisfaction.

REPORT OF CASES

In studying the results of our investigation it has developed that many cases of reduced renal function for uric acid cannot be accounted for from a clinical standpoint by a diagnosis of nephritis. Thus many cases in which a clinical diagnosis of nephritis is not possible, show such a reduction. The fundamental questions for consideration relate to the genesis and significance of such reduced function. It would seem possible that such genesis and significance might be as follows:

Due either to errors in diet or from a variety of perverted body functions existing over a shorter or longer period of time there occurs an excessive supply to the blood of various end products of which uric acid is one. In this way the kidney is put under stress of excessive elimination. Since it is a physiologic law that the body organs in general respond to stimulation, so at first there is probably a period of increased renal function. As time goes on, however, and repeated

TABLE 3.—Uric Acid Retention Due to Varying Causes

No.	Name	Age	Blood Pressure	Cardiac Area Verified by Roentgenograms	Cardiac Murmur	Brachialis	Headache	Vertigo	Vision	Eye Ground	Loss of Weight	Digestive Disturbances	Edema	Albumin	Casts	Blood Uric Acid,* Mg. per 100 C.c.	Urine Uric Acid,* Mg. per 100 C.c.	Renal Concentration	Etiologic Types
1	M. N.	26	90	—	—	—	+	+	+	—	+	+	—	—	—	10.14	35.58	0.96	Endocrine
2	M. S.	33	150	+	—	—	+	+	+	—	+	+	—	+	+	4.4	25.00	1.00	Endocrine
3	J. J. D.	45	126	+	—	—	+	+	+	—	+	+	—	—	—	9.46	74.00	1.1	Uric acid
4	J. J. D.	48	110	+	—	—	+	+	+	—	—	+	—	—	—	6.10	30.00	1.4	Uric acid
5	C. W.	35	120	+	—	—	+	+	+	—	+	+	—	—	—	11.70	57.50	0.51	Poor metabolism
6	L. B.	34	140	+	+	—	+	+	+	+	+	+	—	+	—	15.15	26.40	0.35	Uric acid
7	G. G.	28	90	—	—	—	+	+	+	+	+	+	—	+	—	4.88	53.50	1.8	Poor metabolism
8	H. P. B.	43	90	—	—	—	+	+	+	—	+	+	—	+	—	6.18	45.00	1.00	Poor metabolism
9	N. W.	52	120	+	+	—	+	+	+	+	—	—	—	—	—	7.7	53.50	1.00	Arteriosclerosis
10	T. C.	58	180	+	+	+	+	+	+	+	—	—	—	+	—	6.18	19.70	0.86	Arteriosclerosis
11	R. F. F.	50	130	+	+	+	+	+	+	+	+	+	—	—	—	10.00	40.80	3.4	Arteriosclerosis
12	G. R.	48	128	+	+	+	+	+	+	+	+	+	—	—	—	3.46	15.40	1.00	Def. Dig. function
13	W. R. L.	28	126	—	+	+	+	+	+	+	+	+	—	—	—	3.88	14.42	1.8	Def. Dig. function
14	J. W.	63	150	—	—	+	+	+	—	+	+	+	—	—	—	4.44	37.50	1.3	Pernicious anemia
15	J. L. M.	27	110	—	+	—	+	+	—	—	+	+	—	+	—	8.25	10.50	0.16	Def. Dig. function
16	J. W. P.	38	90	+	+	—	+	+	—	—	—	+	—	—	—	8.08	15.05	0.43	Def. Dig. function
17	S. G.	18	110	+	+	—	+	+	—	—	—	+	—	—	—	6.20	20.40	1.2	Def. Dig. function
18	R. W.	38	140	+	+	—	+	+	—	—	—	+	—	—	—	5.05	40.25	1.9	Def. Dig. function
19	R. P. S.	41	170	—	—	—	—	+	—	—	—	+	—	+	—	5.5	66.00	1.3	Def. Dig. function
																	†		
20	P. W. G.	34	128	+	+	—	+	+	—	—	—	+	—	—	—	6.15	1.08	1.3	Def. Dig. function
21	M. A. F.	44	120	+	+	—	+	—	—	—	—	+	—	—	—	6.50	0.642	1.08	Def. Dig. function
22	G. McG.	53	140	—	—	—	—	—	—	—	—	+	—	—	—	5.75	0.247	0.60	Def. Dig. function
23	J. A.	53	180	+	+	—	—	—	—	—	+	+	—	—	—	5.00	0.592	0.89	Def. Dig. function
24	J. E. B.	43	120	—	—	—	—	—	—	—	—	+	—	—	—	2.52	0.688	2.60	Def. Dig. function
25	B. L.	32	100	—	—	—	—	—	—	—	+	+	—	+	—	3.00	0.435	1.00	Def. Dig. function

* Normal blood uric acid per 100 c.c. 1 to 3 mg.
† All names following are gm. per twenty-four hours.

TABLE 4.—CHRONIC INTERSTITIAL NEPHRITIS

No.	Name	Age	Blood Pressure	Cardiac Area Verified by Roentgenograms	Cardiac Murmur	Brachials	Headache	Vertigo	Vision	Eye Ground	Loss of Weight	Digestive Disturbances	Edema	Albumin	Casts	Blood Uric Acid,* Mg. per 100 C.c.	Urine Uric Acid, Mg. per 100 C.c.	Renal Concentration
1	A. W. A.	49	170	+	—	+	+	+	+	+	—	—	—	—	—	3.16	38.8	6.2
2	A. W. B.	60	170	+	+	+	+	+	+	+	—	+	—	—	—	6.27	22.0	11.2
3	J. B.	65	170	+	+	+	+	+	+	—	—	—	—	+	—	5.06	36.25	1.0
4	H. K.	38	142	—	—	—	+	+	+	—	+	+	—	—	—	7.86	18.72	0.74
5	V. A.	58	154	+	+	+	+	+	+	—	+	+	—	—	—	7.65	51.3	1.8
6	J. J.	51	200	+	+	+	+	+	+	+	+	+	—	+	+	6.79	46.9	6.9
7	H. H.	14	110	—	+	—	+	+	+	—	+	+	—	+	+	4.00	56.00	0.95
8	H. B. D.	38	108	+	+	—	+	+	—	—	+	+	—	—	—	7.12	35.7	5.0
9	M. T.	46	150	+	+	—	+	+	—	—	+	+	—	—	—	2.94	18.75	1.8
10	H. R. M.	61	120	—	—	+	+	+	+	—	+	+	—	—	—	5.74	55.00	1.1
11	R. G. Y.	33	168	—	—	—	+	+	+	—	+	+	—	+	—	9.24	30.87	0.53
12	A. B. H.	33	110	+	+	—	+	+	+	—	+	+	—	—	—	4.58	1.02	0.067
13	B. M.	50	90	+	+	+	+	+	+	+	+	+	—	+	—	4.62	24.00	0.53
14	A. E. M.	52	200	+	+	+	+	+	+	—	+	+	—	+	+	9.15	18.45	0.39
15	G. P.	28	110	—	—	—	+	—	—	—	+	—	—	—	—	1.90	46.6	3.6
16	E. A.	28	110	+	+	—	+	+	—	—	+	+	—	+	—	6.69	23.5	1.3
17	R. F. T.	60	130	+	+	+	+	—	+	—	+	+	—	—	—	10.60	40.80	0.66
18	R. T. S.	45	108	—	—	—	+	—	—	—	+	+	—	—	—	7.86	6.48	0.18
19	C. L.	52	110	—	—	—	—	—	—	—	—	+	—	—	—	5.08	16.30	0.82
20	.....	..	...	..	..	..	+	..	..	..	..	..	..	..	..	....	†	
21	G. R. W.	67	180	+	+	+	+	+	+	—	+	+	—	—	—	6.70	1.27	0.09
22	A. M. H.	5 5	180	+	+	+	+	+	+	+	+	—	—	—	—	6.00	0.45	0.59
23	E. W. H.	51	190	+	+	+	+	+	+	+	+	+	—	+	—	5.55	0.45	0.58
24	J. A. P.	57	150	—	—	+	+	—	—	—	—	+	—	—	—	6.50	2.32	1.4
25	M. B.	58	200	+	+	+	+	+	+	—	+	+	—	+	—	8.10	5.37	1.1
26	E. H.	60	190	+	+	+	—	—	+	—	—	+	—	—	—	5.08	0.386	0.97
27	E. B.	78	200	+	+	+	+	+	+	+	+	+	+	—	—	1.87	0.25	0.50

* Normal blood uric acid per 100 c.c. from 1 to 3 mg.
† All names following are gm. per twenty-four hours.

onslaughts are made on the renal function, such function falters with a concurrent increase of the amount of blood uric acid. Whether such faltering is due to renal anatomic changes and is in reality the initial stage, hitherto unrecognized, of an anatomic nephritis is a question which can only be decided by a considerable number of coincident blood examinations and necropsy investigations. If such should prove true, an anatomical nephritis as it has been understood is a terminal symptomatology of a process of perverted metabolism or dietary errors. We would not be understood as thinking that the above would offer the only explanation of nephritic genesis, for the casual relation of bacterial toxins and other toxic substances to nephritis has been advanced on logical grounds. In such cases it is evident that the primary excitant of the reduced renal function is the local effect on the kidney of these toxic substances.

From these cases it is seen that a reduced renal function for uric acid is present in a variety of conditions other than an anatomic nephritis—the presence or absence of such anatomical nephritis being decided by the conventional methods. If, however, the foregoing reasoning has any basis in fact the discovery of the reduced renal function is of prime importance.

For, from the standpoint of treatment and prognosis the causes of the reduced renal function must be ascertained and corrected.

The discovery of reduced renal uric acid function, therefore, while it does not give by itself a diagnosis of anatomical nephritis, is of much greater value than if such were the case. This is so because it is an early sign of kidney damage, often occurring before a clinical diagnosis of an anatomical nephritis can be made.

The practical application from a clinician's standpoint in treating these cases of low uric acid co-efficients has been by three methods. First, by the reduction of the purin diet; second, by increasing the cardiac function, and, third, by therapeutic methods toward increasing renal elimination. The results obtained were extremely satisfactory at times, sometimes with the use of one method, other times with the use of the second method or by the combination of one or more methods. But other cases failed to show improvement and from a study of these cases we were led to believe that it was due to a second factor of retention in the blood, and it is along this line that our investigations of the second substance or possibly three substances of retention is being made. Thus our studies are not being limited to blood uric acid alone, but at the present time are including other substances of blood chemistry.

HYMENOLEPIS NANA; POSSIBLE CERCOCYSTIS STAGE *

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Since the prediction of Stiles [1] that *Hymenolepis nana* would prove to be a common intestinal parasite in the United States, cases have been reported with increasing frequency, and the indications are that it is the most common tapeworm in many parts of the United States.

Ransom [2] compiled the United States cases up to 1904, reporting twenty-five cases, all but one of the twenty-five having been reported after 1902.

Deaderick,[3] in reporting two new cases in 1910, found that the number had reached thirty-three.

Schloss [4] in the same year reported twenty cases of *Hymenolepis nana* in 280 children examined for intestinal parasites and only five cases of *Tenia saginata.*

Amesse [5] reported one case from Professor Howland's service at Bellevue Hospital.

M. A. Wood [6] reported three cases of *Hymenolepis nana* from Houston, Texas.

Bass and Gage [7] reported fifteen cases out of 577 persons examined in New Orleans, and only three cases of *Tenia saginata.*

H. B. Wood,[8] from records of examinations made during 1911 and the first quarter of 1912 in the state laboratories of the South, found *Hymenolepis nana* in 1,004 of 62,785 persons examined, and only ten cases of *Tenia saginata.*

Deaderick and Thompson [9] stated that during 1911-1913, of 56,543 infections found by the Rockefeller Sanitary Commission, 1,879, or 3.3 per cent., were dwarf tapeworm.

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* This paper was read in abstract by Dr. George Dock at the meeting of the American Society of Tropical Medicine, held at New Orleans, April 27, 1920. In the discussion that followed it was shown that *Tenia saginata* is much less common in some places than it was about thirty years ago.

1. Stiles, C. W.: New York M. J. **77**:877 (Nov. 7) 1903.
2. Ransom, B. H.: U. S. P. H. Service, Hyg. Lab. Bull. **18**: 1904.
3. Deaderick, W. H.: Arch. of Schiffs u. of Trop. Hyg. **14**:21, 1910.
4. Schloss, O.: Am. J. M. Sc. **139**:675, 1910.
5. Amesse, J. W.: Colorado M. **7**:483, 1910.
6. Wood, M. A.: Texas State J. M. **6**:144, 1910.
7. Bass, C. C., and Gage, J. W.: New York M. J. **92**:769 (Oct. 15) 1910.
8. Wood, H. B.: J. A. M. A. **59**:1707 (Nov. 9) 1912.
9. Deaderick, W. H., and Thompson, L.: The Endemic Diseases of the Southern States, p. 503.

Gerber [10] reported the first case from Boston.

Judkins [11] found seventy-one cases among 15,000 people examined throughout Texas.

McNeil [12] reported six cases of *Hymenolepis nana* in the Southern Pacific Hospital in Houston, Texas.

Greil [13] found seventy-five children under twelve years of age in Montgomery, Ala., with dwarf tapeworm infection.

Frey [14] reported that out of 118 cases of parasite infections in the Texas State Orphan Home, 32.6 per cent. were *Hymenolepis nana.*

Van Liere [15] reported one case in twenty foreign students examined at the University of Wisconsin.

Willets [16] reported six cases in the Georgia State Sanitarium.

Lyon,[17] in a study of 477 patients at the Walter Reed General Hospital, found two cases of dwarf tapeworm and five cases of *Tenia saginata.* The latter, however, were diagnosed before being referred to the hospital.

Kofoid and Kornhauser [18] examined the stools of 1,200 American soldiers who had been overseas and of 300 home service men. In the former series there were seven cases of *Hymenolepis nana;* in the latter none.

DeBuys [19] studied 595 children from seven different institutions in New Orleans, and found *Hymenolepis nana* in fifty-five cases, or 9.25 per cent. No other tapeworms were found.

Lucke [20] reported 230 cases of dwarf tapeworm in 35,000 white and black troops at Camp Zachary Taylor, Kentucky. *Tenia saginata* was found twelve times.

Notwithstanding the fact that the parasite has been one of the most frequently encountered, according to the reports of several authors, yet in many localities *Hymenolepis nana* has either not been looked for or has not been found. So far as is known there are no case reports from the region of St. Louis.

The following cases occurred in that city in the B. family, colored. The family came to St. Louis from Mississippi early in 1917. There were eight children, seven of whom were infected with the dwarf

10. Gerber, I.: Boston M. & S. J. **168**:346 (March 6) 1913.
11. Judkins, O. H.: Texas State J. M. **10**:126 (July) 1914.
12. McNeil, H. L.: Southern M. J. **8**:486 (June 1) 1915.
13. Greil, G. J.: Am. J. Dis. Child. **10**:363 (Nov.) 1915.
14. Frey, J. H.: Texas State J. M. **11**:229, 1915.
15. Van Liere, E. J.: J. A. M. A. **67**:1369 (Nov. 4) 1916.
16. Willets, D. J.: Southern M. J. **10**:42 (Jan. 1) 1917.
17. Lyon, M. W.: J. A. M. A. **72**:326 (Feb. 1) 1919.
18. Kofoid, C.; Kornhauser, S., and Plate, J. T.: J. A. M. A. **72**:1721 (June 14) 1919.
19. DeBuys, L. R., and Dwyer, H. L.: Am. J. Dis. Child. **18**:269 (Oct.) 1919.
20. Lucke, B.: Mil. Surgeon **44**:620 (June) 1919.

tapeworm and the eighth with *Ascaris lumbricoides*. The mother had pin worms, the father was free from parasites. The first patient was sent into Barnes Hospital from the out-patient pediatric service; the others were seen at their home.

REPORT OF CASES

CASE 1.—F. B., male, aged 5 years, born in Mississippi. Entered Barnes Hospital, Jan. 19, 1920, on account of severe diarrhea. In October, 1919, he began having numerous loose mucoid stools with blood, sometimes bright red, sometimes tarry. Considerable straining at stool, no vomiting or pain; appetite greatly increased. Physical examination showed moderate distention of abdomen. Red blood cells, 4,160,000; hemoglobin, 80 per cent.; eosinophilia, 7 per cent. Stool was liquid, light yellow, with mucus, benzidin positive; very many ova of *Hymenolepis nana*.

Treated with male fern January 21; no worms recovered. Stools egg free up to January 28, an interval of seven days. Treatment repeated February 10 and at least one thousand *Hymenolepis nana* worms were obtained. Stools were egg free up to February 23, when many small circular bodies from 5 to 15 microns in diameter were observed. February 24, thymol, 15 grains, was given, and one worm was found. Stools egg free to March 15, an interval of eleven days. March 19 male fern was given but vomited, and March 20 another dose was given through a tube. Many tags of tissue containing heads of dwarf tapeworm embedded within the tissue were found. No whole worms or segments were observed. Stools egg free to March 29, an interval of ten days. Discharged from hospital April 4. Stool on discharge, soft, brown, little mucus, no blood; dwarf tapeworm eggs, from 1 to 3 per low power field. April 18, condition good; stool as on April 4. May 28, patient had had measles in interim and was very anemic; passing considerable blood in stools. Stool was soft, yellow and blood streaked and contained large numbers of *Hymenolepis nana* eggs.

CASE 2.—J. B., male, aged 1 year 7 months, born in St. Louis. February 22, no symptoms; physical examination negative; stools hard, brown, few *Hymenolepis nana* eggs found; eosinophils, 3 per cent.

CASE 3.—E. B., female, aged 3 years, born in Mississippi as were all the others. All through January had four or five loose stools a day. For the past four months has frequently had similar symptoms. Complains of abdominal pain during these periods. February 22, stool gray-brown, much mucus, many *Hymenolepis nana* eggs; eosinophils, 6 per cent.

CASE 4.—A. B., female, aged 8 years. At 3 years passed twenty-three ascarides (?). Occasional short attacks of diarrhea for past four months, with abdominal pain. Nausea and vomiting, frequently with dizziness. Patient is anemic and drowsy. Hemoglobin, 70 per cent.; red blood cells, 3,500,000; white blood cells, 9,000; eosinophils, 9 per cent. February 22, stool soft, gray, with considerable mucus. Very many *Hymenolepis nana* eggs. March 29, course of male fern. Several thousand *Hymenolepis nana* worms obtained. April 17, general condition good; no symptoms since treatment. No *Hymenolepis nana* eggs found in stools.

CASE 5.—A. B., male, aged 8 years. Complains of frequent frontal headaches. Examination negative; eosinophils, 3 per cent. February 22, stool normal; no eggs found in preparations. March 27, very many *Hymenolepis nana* eggs.

CASE 6.—R. B., male, aged 9 years. No symptoms; physical examination negative; eosinophils, 2 per cent.; stool normal; few *Hymenolepis nana* eggs.

CASE 7.—M. B., female, aged 11 years. For the past year has had attacks of diarrhea, the last one early in February. Frequently has tenderness in abdomen and gets dizzy at times. Recalls no symptoms previous to past year.

Patient anemic; has slight abdominal tenderness. February 22, stool normal; very many *Hymenolepis nana* eggs. Eosinophils, 7 per cent.; red blood cells, 3,800,000; white blood cells, 8,000. March 29, course of male fern; at least one thousand worms obtained. April 17, few *Hymenolepis nana* eggs found. No symptoms since treatment.

The morphology of the worms does not differ essentially in any respect from the usual descriptions. Specimens were sent to Dr. C. W. Stiles, who kindly reported that he identified the worms as *Hymenolepis nana*, the eggs belonging to the same.

In Case 1, the large number of very young specimens found, the smallest being 3 x 0.15 mm., is noteworthy. The largest specimens were 15 mm. in length, the maximum width 0.6 mm. Senna, cited by Ransom,[2] noticed in feces containing *Hymenolepis nana* eggs many small rounded bodies measuring from 5 to 30 microns in diameter. The smaller of these were homogenous, with a thin membrane, while the larger were more granular and tended to become oval, with thicker membrane. Senna thought these might be eggs in the course of development which had prematurely escaped from the uterus, but finding similar bodies in two cases in which he could not demonstrate the presence of *Hymenolepis nana*, he was left in doubt as to their true nature. In Case 1, of this series, thirteen days after the second course of male fern and before the eggs had reappeared in the stools, there were found a large number of homogenous structures from 5 to 15 microns in diameter, bounded by a thin membrane. The next day, for the first time, *Hymenolepis nana* eggs were observed, with fewer of the smaller structures. These were not observed again.

SYMPTOMS

Of the seven cases four had definite symptoms, which, in the absence of other factors, may be attributed to the dwarf tapeworm. Two of the children had no symptoms and in both cases very few eggs were found. One patient with a heavy infection had long-standing headache, which may possibly have been due to the parasites. The most frequent symptoms were abdominal pain, or tenderness, and diarrhea, found in four cases. There were anemia, dizziness and headache in two cases, increased appetite in one case. The presence of blood in Case 1 is interesting. Innes, cited by Ransom, in reporting a necropsy on a case, states that he found bloody extravasations on the mucous membrane of the ileum, which may have been the points of attachment of the tapeworms lying free in the intestine.

Eosinophilia, according to Schloss [21] and others a constant finding in cases showing symptoms, ranged from 2 to 9 per cent. In the children with definite symptoms it was from 6 to 9 per cent., while in the others it was from 2 to 3 per cent.

21. Schloss, O.: Arch. Pediat. **27**:100 (Feb.) 1910.

Treatment was instituted in three cases, and an apparent amelioration of symptoms occurred in all. Case 1, after four courses of treatment, still showed a few eggs in the stool on the day of the patient's discharge. Subsequently, after an attack of measles, his intestinal symptoms reappeared. In Case 4 apparently complete cure was obtained after one course of male fern. In Case 7 few eggs showed after one course of treatment.

MODE OF INFECTION

The development as well as the manner of infection of the dwarf tapeworm is unknown. There is a form of tapeworm common in rats, the *Hymenolepis murina* of Dujardin, which morphologically is very similar to *Hymenolepis nana*. Grassi,[2] quoted by Ransom, found this parasite common in rats in Catania, where also the dwarf tapeworm was common. In a series of carefully conducted experiments, he showed that the feeding of mature segments of *Hymenolepis murina* to rats was followed by infection with this tapeworm. The eggs liberate six-hooked embryos in the small intestine, which enter the villi of the last part of the ileum and there become transformed into cercocysts. The position of each cercocyst is in the dilated central lymphatic cavity of the villus. Subsequently, the cercocyst leaves its position in the villus and becomes changed to the adult worm, and is attached to the epithelium of the intestines. Just how the latter stage ensues is not noted. Joyeux [22] repeated these experiments successfully.

Grassi, Lutz and Ransom consider *Hymenolepis nana* of man identical with the rat hymenolepis, but Braun, Loos and others doubt this. Grassi, Loos, Stiles,[23] Schnell,[24] and Joyeux [25] were unable to transmit the infection to rats and mice. Stiles states that the "form from rodents is entitled to at least subspecific rank." Castellani [26] states that the dwarf tapeworm of man is probably distinct from the rat type.

Minchin and Nicholl, and later Johnston,[27] state that they found cysticercoids of *Hymenolepis murina* in the body cavity of *Xenopysylla cheopis* and *Ceratophyllus fasciatus*, but Joyeux states that he was unable to transmit the infection experimentally in these and other fleas.

Grassi fed worms and eggs of both types to eight indidivuals and in only one case were adult tapeworms found. In this case, a boy

22. Joyeux, C.: Bull. Soc. Path. Exot. **9**:578, 1916.
23. Stiles, C. W.: Osler's Modern Medicine **2**:252.
24. Schnell, W.: Centralbl. f. Bakteriol., Abt. **82**:304 (Nov. 11) 1918.
25. Joyeux, C.: Bull. Soc. Path. Exot. **12**:228 (May 14) 1919.
26. Castellani and Chalmers: Manual of Tropical Medicine, Ed. 3, p. 610.
27. Johnston, T. H.: Proc. Roy. Soc. Queensland **24**:63, 1913.

of 5 began to pass eggs fifteen days after ingesting several segments of the rat hymenolepis and later expelled fifty worms on treatment. In another instance a boy who was previously free from *Hymenolepis nana* was infected after a month during which time he was collecting the feces of an infected patient. Grassi himself raises objections to these cases because of the high incidence of *Hymenolepis nana* in the locality and because worms may be present, although the stools are egg free.

The experiments of Grassi, together with the failure to find an intermediate host, have led to the assumption that the mode of infection is direct, and that man himself, like the rat, may be the intermediate host. Stiles accepts this possibility. As possible evidence of direct infection, Grassi cites cases of dwarf tapeworm infection in several individuals, previously egg free, in whose families there were known to be cases of *Hymenolepis nana*. Schloss [21] cites two similar cases in his series. The frequent findings of infection in several members of the same household would also point to the possibility of a direct infection. Thus the Hygienic Laboratory staff [28] found five cases in the same ward on an insane asylum. Magnenat, quoted by Stiles,[1] reported four cases in the same family. In all but one of Schloss' cases several members of the same family were infected. Carpenter,[29] in discussing DeBuys' paper before the American Pediatric Society, described an epidemic of *Hymenolepis nana* in a foundling asylum in Philadelphia and thinks the infection was direct. Rats examined by him were negative.

I attempted to determine the mode of infection of the dwarf tapeworm. The feeding of eggs and mature segments to six rats and six mice was negative. Attempts to incubate the eggs following their mixture with artificial gastric juice were also negative.

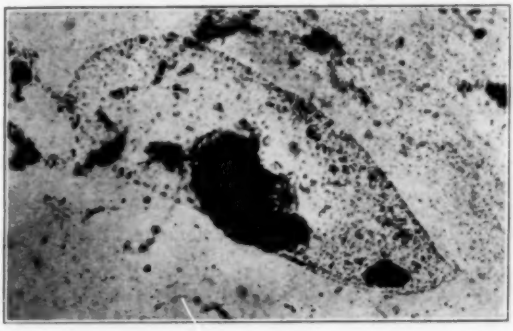
The house of family B was carefully inspected and found to be in a very unsanitary condition. The children played around the floor a great deal so that chances for coprophagia were very favorable. There were no rats. Several bedbugs were examined and were negative for cysticerci. Several lice from the head of Case 4 were also negative.

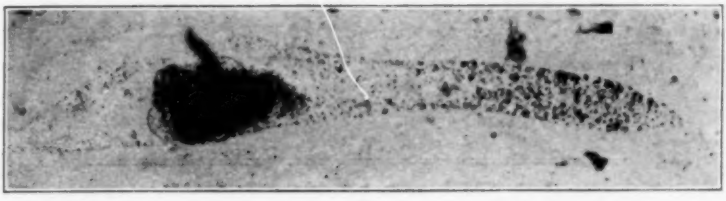
There was an old pet dog with which the children were very intimate. A stool from the dog, obtained while the dog was at the B. home, showed several eggs, which were exactly similar to eggs of *Hymenolepis nana*, the characteristic filaments of the latter being present. There were also present trichomonads and eggs of *Dipylidium caninum* and *Tenia serrata*. The dog was removed from the

28. Stiles and Garrison: U. S. P. H. S., Hyg. Lab. Bull. **28**: 1906.
29. Carpenter, H. C.: Arch. Pediat. **36**:379 (June) 1919.

home and daily examinations of stools were made, but hymenolepis eggs could no longer be found. Two weeks later a necropsy was made and *Dipylidium caninum* and *Tenia serrata*, but no *Hymenolepis nana* were recovered.

The presence of the parasite in all but the oldest boy, age 12, is of interest. In Case 5, no eggs were observed in the specimen of February 22 after twelve examinations, while on March 27 the stool was loaded with them. The difficulty of ridding the host of all the worms is a striking feature. It has been stated by Ransom and other authors that one course of treatment is frequently not sufficient. In two of my three cases which were treated eggs showed subsequently. In Case





Figs. 1 and 2.—Head of dwarf tapeworm surrounded by saclike structure. Head shows some details.

1 four courses of treatment were given and at the end of these there were still many eggs present. The large number of very young forms following the second treatment was striking.

Following the last administration of the vermifuge, the full dose being repeated next day because of the vomiting of the male fern, there were passed a number of mucoid looking tags of tissue, but no worms. The largest of these pieces were 20 x 10 mm. Each had from two to eight heads of *Hymenolepis nana* studded throughout it, but no segments could be found. Microscopically, there were observed the rostellum, a row of hooklets, four suckers and a small caudal append-

age. Surrounding each head there could be seen in the unstained specimen a definitely clear area. In the stained specimen, there was seen microscopically a saclike structure completely surrounding each head. The shape of the structures varied, some being elliptical, others ovoid, still others approaching a spherical shape. Grossly, both the head and the sac could be seen in the stained tissue. It does not seem likely that the adult worm could develop within the saclike structure.

Figures 1 and 2 show the relative size and shape of the head and sac. The tissues were stained several times with hematoxylin-eosin and also with methylene blue, but no nuclei could be seen. I felt that the structures might possibly represent the cercocyst stage of the dwarf tapeworm, thus indicating that man himself may be the intermediate host. The occurrence of autoinfection is proved by the large number of worms present and the difficulty in getting completely rid of them.

SUMMARY

1. A review of the literature shows that the dwarf tapeworm is the most common tapeworm in many parts of the United States.

2. The first cases of *Hymenolepis nana* from St. Louis are reported.

3. A possible cercocyst stage of *Hymenolepis nana* is demonstrated in man.

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